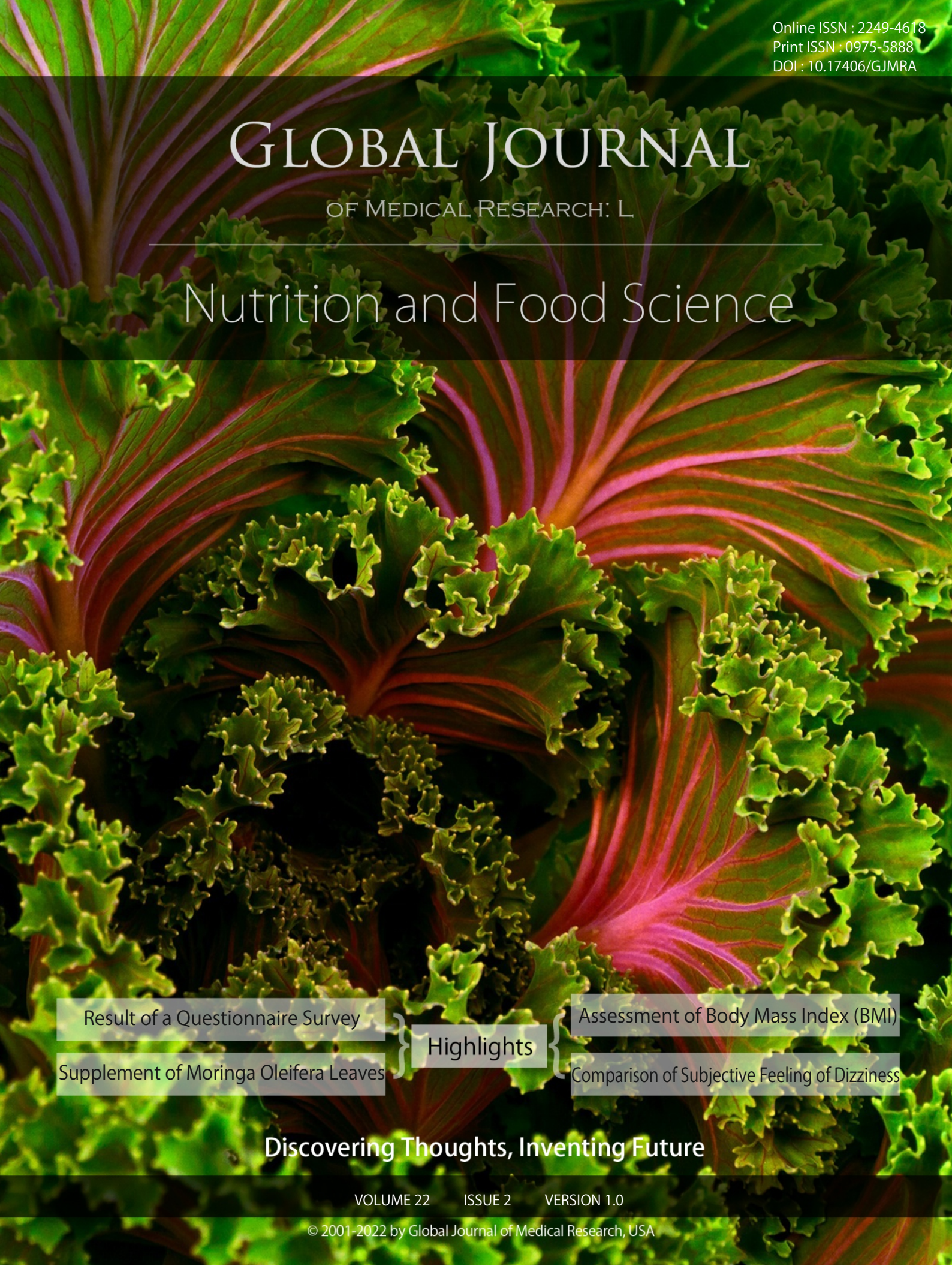
Online ISSN : 2249-4618
Print ISSN : 0975-5888
DOI : 10.17406/GJMRA

Global Journal

of Medical Research: L

Nutrition and Food Science



Result of a Questionnaire Survey

Supplement of Moringa Oleifera Leaves

Highlights

Assessment of Body Mass Index (BMI)

Comparison of Subjective Feeling of Dizziness

Discovering Thoughts, Inventing Future

VOLUME 22 ISSUE 2 VERSION 1.0

Global Journal of Medical Research: L
Nutrition & Food Science

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Volume 22 Issue 2 (Ver. 1.0)

Open Association of Research Society

This is a special issue published in version 1.0 of "Global Journal of Medical Research." By Global Journals Inc.

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Offset Typesetting

Global Journals Incorporated
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Nutrition & Food Science
Volume 22 Issue 2 Version 1.0 Year 2022
Type: Double Blind Peer Reviewed International Research Journal
Publisher: Global Journals Inc. (USA)
Online ISSN: 2249-4618 & Print ISSN: 0975-5888

Comparison of Subjective Feeling of Dizziness and Simple Taste/Olfactory Test Results in Elderly People (Over 60 Years Old)

By Naomi Katayama & Shoko Kondo

Nagoya Womens University

Abstract- For a long time, the author has been involved in taste and smell with Yakumo Town (rural) residents in Hokkaido and Nagoya-City (urban) in Aichi Prefecture. Participants answered a self-administered questionnaire, and then took a simple salty taste test and a simple olfactory test.

However, until now, the author has not been able to compare the results of a questionnaire survey of Yakumo Town, Hokkaido, and residents of Nagoya City, Aichi Prefecture. Therefore, this time, we will report the results.

201 residents of Yakumo Town (95 men, 106 women: 2019 data) and 55 residents of Nagoya City 24 males and 31 females: 2022 data) participated in the examination.

A self-reported questionnaire was given to the participants to determine the presence or absence of dizziness (1, no dizziness, 2. dizziness, and 3. dizziness all the time).

Keywords: dizziness, taste, olfactory, questionnaire survey.

GJMR-L Classification: DDC Code: 004.7 LCC Code: QA76.889

Strictly as per the compliance and regulations of:

Comparison of Subjective Feeling of Dizziness and Simple Taste/Olfactory Test Results in Elderly People (Over 60 Years Old)

Comparison between Residents of Yakumo Town, Hokkaido and Residents of Nagoya City, Aichi Prefecture

Naomi Katayama [α] & Shoko Kondo [σ]

Abstract- For a long time, the author has been involved in taste and smell with Yakumo Town (rural) residents in Hokkaido and Nagoya-City (urban) in Aichi Prefecture. Participants answered a self-administered questionnaire, and then took a simple salty taste test and a simple olfactory test.

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A self-reported questionnaire was given to the participants to determine the presence or absence of dizziness (1, no dizziness, 2. dizziness, and 3. dizziness all the time).

In addition, the participants were given a simple salty taste test (Solceive: manufactured by Advantech), andan olfactory test (smell test: Daiichi Yakuhin Kogyo Co., Ltd.) was performed.

In addition, participants filled in a self-administered questionnaire about their physical conditions (,age, sex, height, weight, systolic blood pressure, and diastolic blood pressure).

As a result, the subjective feeling of dizziness was statistically significantly higher in Nagoya City residents than in Yakumo Town residents (P=0.044*).

In addition, the subjective sense of salty taste and smell was statistically significantly worse in Yakumo Town residents than in Nagoya-shi residents (Salt taste P=0.027*Olfactory P=0.017*).

However, when the results of salty taste and olfactory tests were conducted on the residents of Nagoya City and Yakumo Town, there was no statistically significant difference (salty taste test results P = 0.614, Olfactory test result P=0.052).

Regarding the subjective feeling of dizziness, in the future, we will conduct actual measurements of the sway of the center of gravity using Stabilometer for both residents.

We believe that it is necessary to obtain definite results.

In a self-administered questionnaire sauvery, participants in Yakumo Town answered that it was not well to distinguish between the smell and taste.

We need to ask more detailed questions about the participants' dietary habits in the future.

I think that life survey is necessary.

Keywords: dizziness, taste, olfactory, questionnaire survey.

I. Introduction

Since 2005, I have conducted a simple taste/olfactory test and a self-administered questionnaire at the health checkup for residents of Yakumo Town, Hokkaido[1-11].

Similarly, a simple taste/olfactory test and a self-administered questionnaire survey were conducted at a health class for residents of Nagoya City[12-20].

However, until now, no comparison has been made between the two regions. Therefore, we compared the results of these two regions this time.

Residents of Yakumo Town (FY2019) and Nagoya City (FY2022) were asked to feel dizziness, taste, and olfaction by using a self-administered questionnaire.

And participants also took simple salty taste test and a simple olfactory test.

At the same time, primary data such as age, sex, height, weight, systolic blood pressure, and diastolic blood pressure were obtained.

A questionnaire survey was also conducted on subjective dizziness.

Feeling dizzy (light-headedness, fluffiness) due to changes in the amount and contents of food associated with the decline in taste and smell[21] related to Yakumo Town whaich is located in the south part of Hokkaido island in the northern part of Japan.

There is a little population movement, and the population is settled.

On the other hand, Nagoya City is located almost in the center of Japan, between Tokyo and Osaka.

Because it is a large city, there are various occupations, and the population movement is rapid.

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This study has so far been a self-reported survey of taste, smell, and different living conditions in health checkups for residents of Yakumo Town, Hokkaido.

I've been researching it with a questionnaire, but I haven't made a comparison with other places.

Therefore, in this study, we decided to compare the data obtained from urban and rural paticipants.

II. Materials and Methods

Two houndred one people in Yakumo Town, Hokkaido (106 women, 95 men: 2019 data) and 55 people in Nagoya City, Aichi Prefecture (31 women, 24 men, 2022 data) were compared.

Dizziness was also included in the self-administerd questionnaire survey of the participants. We asked the following questions.1.not dizzy, 2. sometimes, 3. always. Participants circles the items that apply.

In addition, a simple taste test (salty taste: Soluseive: manufactured by Advantech) and a simple olfactory test (smell test) were performed.

Ick: manufactured by Daiichi Yakuhin Sangyo Co., Ltd.) was used to obtain the test results.

In addition, praimary data such as age, sex, height, weight, systolic blood pressure, and diastolic blood pressure were obtained.

Other self-administered questionnaire items included the subjective sense of taste, smell, salivary flow, and use of eating out. We also investigated the frequency and usual seasoning. (See Table 1):

The method of the simple taste test[22] and the method of the simple olfactory test[23] followed the specifications.

Table 1. Questionnaire question content

	1	2	3	4	5	6
Sex	Male	Female				
dizziness	None	Sometimes	Common			
Saliva	Good	normal	bad			
olfactory	Good	normal	bad			
taste	Good	normal	bad			
requency of eating out	Daily	4-5 times / week	2-3 times / weed	one time / week	2-3 times / month	Rarely used
teste of everyday meals	Strong seasoned	Slightly seasoned	slightly lightly seasoned	lightly seasoned		

a) Ethical review board

This study conducted with the approval of the Ethical Review Board (Nagoya women's University Ethics Committee: "hitowomochiitakennkyuunikann suruiinnkai"). The approval number is 2019-26.

III. Results

There were no regional differences in sex (see Table 2) and hight (see Table 4) in the participants' physical data.

Weight (see Table 5), systolic blood pressure (see Table 6), and diastolic blood pressure (see Table 7) were eight higher in Nagoya.

It was statistically significantly lower than Kumocho.

Yakumo town has 201 people (see Table 3) average ± SD value of 68.7 ± 6.0 years old Nagoya city has 55 people 74.9±7.1.

Table 2. Comparison of participant gender data

	N	City n=55	Y	Town n=201
Average value		1.564		1.527
Standard deviation		0.501		0.5
F-test		P=0.484		
Unpaired Student-t test		P=0.634		
Mann- Whitney test				

Table 3. Comparison of participant age data

	N	City n=55	Y	Town n=201
Average value		74.855		68.687
Standard deviation		7.083		5.956
F-test		P=0.045*		
Unpaired Student-t test				
Mann- Whitney test		P=0.0001**		

Table 4. Comparison of participant height data

	N	City n=55	Y	Town n=201
Average value		158.838		159.361
Standard deviation		7.468		8.562
F-test		P=0.080		
Unpaired Student-t test		P=0.672		
Mann- Whitney test				

Table 5. Comparison of participant weight data

	N	City n=55	Y	Town n=201
Average value		55.62		60.618
Standard deviation		8.824		11.476
F-test		P=0.005**		
Unpaired Student-t test				
Mann- Whitney test		P=0.005**		

Table 6. Comparison of participant systolic blood pressure data

	N	City n=55	Y	Town n=201
Average value		124.587		139.731
Standard deviation		13.621		20.518
F-test		P=0.0001**		
Unpaired Student-t test				
Mann- Whitney test		P=0.0001**		

Table 7. Comparison of participant diastolic blood pressure data

	N	City n=55	Y	Town n=201
Average value		71.116		78.303
Standard deviation		8.393		12.239
F-test		P=0.0001**		
Unpaired Student-t test				
Mann- Whitney test		P=0.007**		

The average ± SD value for subjective dizziness (see Table 8) was 1.379 ± 0.592 in Yakumo Town, and 1.379 ± 0.592 in Nagoya City was 1.211 ± 0.546.

This result was P = 0.044* in the Mann-Whitney test, and was statistically superior to the elderly in Nagoya City.

The results showed that the subjects had dizziness subjectively.

Table 8. Comparison of participant aware feeling of dizziness data

	N City n=55	Y Town n=201
Average value	1.379	1.211
Standard deviation	0.592	0.546
F-test	P=0.005**	
Unpaired Student-t test		
Mann- Whitney test	P=0.044*	

The subjective taste (see Table 9) has a mean ± SD value of 0.1.607 ± 0.538 in Yakumo and 1.426±0.49 in Nagoya.

This result was P = 0.027* in the Unpaired Student-t test, showing a statistically significant.

From this result, it was found that the participants in rural areas subjectively felt that the taste was difficult to understand compared to those in the urban areas.

Table 9. Comparison of participant aware taste data

	N City n=55	Y Town n=201
Average value	1.426	1.607
Standard deviation	0.49	0.538
F-test	P=0.228	
Unpaired Student-t test	P=0.027*	
Mann- Whitney test		

The subjective sense of smell (see Table 10) was 0.701 ± 0.539 in Yakumo Town, and 0.150±0.575 in Nagoya.

From this result, P = 0.017* in the Unpaired Student t-test, which is statistically significant for Yakumo Town.

The results showed that older adults subjectively feel that smell is difficult to understand.

Table 10. Comparison of participant aware olfactory data

	N City n=55	Y Town n=201
Average value	1.5	1.701
Standard deviation	0.575	0.539
F-test	P=0.261	
Unpaired Student-t test	P=0.017*	
Mann- Whitney test		

The subjective saliva output (see Table 11) has a mean ± SD value of 0.781 ± 0.000 in Yakumo Town, and1.773 ±0. 000 in Nagoya City.

This result was P = 0.139 in the Unpaired Student's t-test, and there was no statistically significant difference.

Table 11. Comparison of participant aware saliva data

	N City n=55	Y Town n=201
Average value	1.673	1.781
Standard deviation	0.511	0.471
F-test	P=0.209	
Unpaired Student-t test	P=0.139	
Mann- Whitney test		

The average ± SD value for the frequency of eating out (see Table 12) is 5.095±1.037 in Yakumo Town and4.455 ±1.424 in Nagoya City.

This result was P=0.004** in the Mann-Whitney test, indicating a statistically significant.

The results showed that those with the high frequency of eating out had a high frequency of eating out.

Table 12. Comparison of participant frequency of eating out data

	N City n=55	Y Town n=201
Average value	4.455	5.095
Standard deviation	1424	1.037
F-test	P=0.001**	
Unpaired Student-t test		
Mann- Whitney test	P=0.004**	

The seasoning of ordinary meals (see Table 13) has an average ± SD value of 2.542 ± 0.734 in Yakumo Town and 2.704 ± 0.924 in Nagoya City.

This result was P = 0.155 by the Mann-Whitney test, and there was no statistically significant difference.

Table 13.Comparison of participant teste of everyday meals data

	N City n=55	Y Town n=201
Average value	2.704	2.542
Standard deviation	0.924	0.734
F-test	P=0.012*	
Unpaired Student-t test		
Mann- Whitney test	P=0.155	

The results of the simple salty taste (see Table 14) are mean ±SD values of 0.89 ±0.387 in Yakumo Town and 0.86±0.389 in Nagoya City.

This result was P=0.614 in the Unpaired Student's t-test, and there was no statistically significant difference.

Table 14.Comparison of participant taste test data

	N	City n=55	Y	Town	n=201
Average value		0.86		0.89	
Standard deviation		0.389		0.387	
F-test			P=0.491		
Unpaired Student-t test			P=0.614		
Mann- Whitney test					

The results of the olfactory test (see Table 15) are average ±SD values of 7.348 ± 3.007 in Yakumo and 6.455 ±3.310 in Nagoya.

This result was P = 0.0.052 in the Unpaired Student's t-test, and there was no statistically significant difference.

Table 15 . Comparison of participant olfactory test data

	N	City n=55	Y	Town	n=201
Average value		6.455		7.368	
Standard deviation		3.31		3.007	
F-test			P=0.172		
Unpaired Student-t test			P=0.052		
Mann- Whitney test					

IV. Discussion

For primary data (gender, age, height, weight, systolic blood pressure, diastolic blood pressure), participants were statistically significantly older and underweight than rural participants.

The average value of blood pressure was within the normal renge for both Nagoya data and Yakumo data. However, the Nagoya data was statistically significantly lower than the Yakkumo data.

In addition, there were regional differences in subjective dizziness in this survey.

Urban participants said they were statistically significantly dizzier than country participants.

However, there were no regional differences in the salty taste test results.

And also, there was no regional difference in the olfactory test results in the present data.

However, the P-value after statistical processing was P=0.052, so if we increased the data for urban residents, there was a possibility that there would be a statistically significant difference in the olfactory test results.

The frequency of eating out was statistically significantly higher among participants in urban areas. Still, there was no significant difference between the two regions regarding the seasoning of things. Research results on the relationship between salty test results [24-27] and blood pressure[28] have also been reported, so that future studies, we will investigate the association between dietary habits and blood pressure. It is necessary to investigate this in more detail.

Changes due to age[29] and association with Alzheimer's dementia[30] results such as application to patients[31] have been presented. We think it will be important to investigate resional differences in Japan in the future.

We will continue to do research and collect more data in the future, and not only subjective feelings of dizziness but also stabilization tests by using Stabilometer.

We also believe that a detailed questionnaire survey on dietary habits is necessary.

V. Conclusion

Urban participants seid they were statistically significantly dizzier than country participants. However, there were no regional differences in the results of the salty taste test results. And also, there was no regional difference in the olfactory test results in the present data. However, the P-value after statistica processing was P=0.052, so if we increased the data for urban residents, there was a possibility that there would be a statistically significant difference in the olfactory test results. We look forward to future results.

Acknowledgements

This research was partially supported by the research aid of Chojuiryokenkyukaihatsuhi, 2022 (30-14, Hirokazu Suzuki) and Japanese Society of Taste Technology, 2021 (Naomi Katayam) and the Ministry of Education, Science, Sports and Culture, Grant-in Aid for Scientific Research(C), 2020-2022 (20K02372, Naomi Katayam). We would like to express my deepest gratitude here.

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Global Journal of Medical Research: L
Nutrition & Food Science
Volume 22 Issue 2 Version 1.0 Year 2022
Type: Double Blind Peer Reviewed International Research Journal
Publisher: Global Journals Inc. (USA)
Online ISSN: 2249-4618 & Print ISSN: 0975-5888

Antihyperlipidemic Property of a Dietary Supplement of *Moringa Oleifera* Leaves and *Pleurotus Ostreatus* in Wistar Rats Stressed by Combination of Ethanol-Paracetamol

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Abstract- High amounts of triglycerides and cholesterol in the blood result in the metabolic condition known as hyperlipidemia. There is currently no specific therapy to reduce the effects of this disorder. In underdeveloped nations, metabolic diseases are treated using *Moringa oleifera* and *Pleurotus ostreatus*. Both the nutritional and therapeutic benefits of these two plants are frequently utilized.

Purpose: This study aimsto investigate the antihyperlipidemic property of dietary supplement of *Moringa oleifera* leaves and *Pleurotus ostreatus* in wistar rats.

Materials and methods: A variety of mushroom species were produced in the Mushroom Biotechnology Laboratory, and M. oleifera was developed in the university's botanical garden in Dakar, Senegal.

Keywords: moringa oleifera, pleurotus ostreatus, dietary supplement, antihyperlipidemic, oxidative stress.

GJMR-L Classification: DDC Code: 574.192 LCC Code: QP514.2

Strictly as per the compliance and regulations of:

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Fatou Corka Kane [α], Simo Nemg Fredy Brice [σ], Moundipa F. Paul [ρ] & Wilfred F. Mbacham [ω]

Abstract- High amounts of triglycerides and cholesterol in the blood result in the metabolic condition known as hyperlipidemia. There is currently no specific therapy to reduce the effects of this disorder. In underdeveloped nations, metabolic diseases are treated using *Moringa oleifera* and *Pleurotus ostreatus*. Both the nutritional and therapeutic benefits of these two plants are frequently utilized.

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Materials and methods: A variety of mushroom species were produced in the Mushroom Biotechnology Laboratory, and M. oleifera was developed in the university's botanical garden in Dakar, Senegal. In this study, the extract of these two plants, designated FMP16, was used to treat rats that had been exposed to oxidative stress caused by the combination of ethanol and paracetamol as follows: control (TG), stressed (TP), ethanol-paracetamol treated groups (D1P- D2P- D3P), which received three doses of the supplement at 500 mg/kg, 1000 mg/kg, and 1500 mg/kg, followed by ethanol in five sequential doses of 2 g. To measure: oxidative stress parameters, total plasma cholesterol, triglycerides, low-density lipoprotein (LDL Cholesterol), and high-density lipoprotein (HDL Cholesterol), blood and liver samples were collected.

Results: According to findings, giving rats a meal consisting of a 2:1 ratio of *Moringa oleifera* and *Pleurotus ostreatus* lowered plasma levels of total cholesterol, triglycerides, and low-density lipoprotein (LDL). Compared to TG, it decreased the LDL cholesterol of D1P, D2P, and D3P by 39%, 30%, and 38%, respectively. D2P's SGPT and SGOT concentrations were also decreased by 29% and 28%, respectively, compared to TP. The dosage of 1000 mg/kg would be the most suitable for liver damage.

Conclusion: According to the results of the current study, taking *M. oleifera* leaves and *P. ostreatus* supplements may have health benefits, at least because they affect the lipid profile and liver damage in stressed rats.

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I. Introduction

Alcoholism and other serious health issues are brought on by excessive alcohol usage, including alcoholic liver damage (ALD). Alcoholism has been linked to several illnesses, and it is currently one of the most challenging health issues with substantial medical, social, and economic repercussions. (Pari and Karthikesan, 2001; Sivaraj *et al*., 2010). Alcohol abuse leads to significant illnesses such as hyperglycemia, cirrhosis, cardiovascular disease, pancreatic inflammation, and alcoholic fatty liver. (Ponnappa et al., 2000). Oxidative stress is one of the elements that are crucial in numerous pathways of alcohol-induced harm. The creation of ROS in our bodies is abnormally increased by our unhealthy eating habits and our way of life (smoking, drinking, obesity, and strenuous activity). When organisms experience oxidative stress brought on by free radical damage, antioxidants aid in coping. Antioxidant defenses come from two different sources: the diet, which includes fruits and vegetables, which are rich in vitamins C and E, carotenoids, ubiquinone, polyphenols, and lipoic acid. The other is endogenous and is made up of proteins, enzymes, or tiny molecules suchas glutathione, uric acid, superoxide dismutase, and glutathione peroxidase (ferritin, transferrin, etc.). Additionally, some elements that are significant cofactors include selenium, copper, and zinc. (Pincemail *et al*., 2009).

We were particularly interested in the plant *Moringa oleifera* and the edible fungus *Pleurotus ostreatus* because they contain significant antioxidant content. The plant *Moringa oleifera* Lam. (Moringaceae), also known as Nebeday in Senegal, is of Indian ancestry and is now common throughout Asia and Africa. The leaves are utilized in traditional African medicine and are commonly consumed as a legume. They are an excellent source of protein (19–35% dry matter) (Kane *et al*., 2017; Makkar *et al*., (1996); Abou-Elezz *et al*., (2012) and are rich in metabolizable energy (2273–2978kcal/kg DM) (Makkar *et al*., (1996); Olugbemi *et al*., 2010). They are also rich in vitamins (A, B, C, and E), minerals (0.6–

11.2% dry Matter). The South African ecotype of the plant has been observed to contain 19.3% crude protein (Moyo., 2011). Traditional Chinese medicine uses *M. oleifera* leaves to treat diabetes, headaches, fever, and malnourishment (Ndong *et al*., 2007; Kerrarho., 1994).

Preview studies have shown the health and nutritional interest of edible mushrooms (Zhang *et al*., 2016; Alam *et al*., 2008; Pornariya and Kanok, 2009). *P. ostreatus* has been demonstrated for its antitumor effects, antioxidant properties, antihyperlipidemic effects and antidiabetic effects, (Zhang *et al*., 2016; Abrams *et al.,* 2011; Alam *et al*., 2008; Elmastas *et al*., 2007; Jayakumar *et al*., 2007; Jayakumar *et al*., 2006). A daily intake of 15 g of dried oyster mushrooms would have an anti-hyperlipidemic effect on the subjects, it would also cover up to 50% of the recommended daily intakes of macronutrients and minerals, according to research on *Pleurotus ostreatus* nutritional value and antihyperlipidemic effects on HIV-positive individuals taking ARVs (Abrams *et al*., 2011, Alam *et al*., 2008; Manzi *et al*., 2001; Alam *et al*., 2009; Manzi *et al*., 1999, Kane *et al*., 2017).

Given the rich nutrient, phytochemical, and organoleptic potential of *M.oleifera* and *P.ostreatus*, we designed this study to determine the antihyperlipidemic effect of *Moringa oleifera* leaves and *Pleurotus ostreatus* in Wistar rats stressed by a combination of Ethanol-paracetamol. In this paper, wewill code the dietary supplement by FMP16.

II. Materials and Methods

a) *Plant Material and Preparation of the mixture of Leaves from M. oleifera and P. ostreatus*

The fresh leaves of *M. oleifera* were harvested at the botanical garden of the University Cheikh Anta Diop (UCAD) of Dakar, Senegal and identified at the botanical department (UCAD).The leaves were cleaned immediately after harvest, cut into small pieces, and dried in the shade for two weeks. The dried material was ground into a powder using a manual homogenizer. *P. ostreatus* were obtained by cultivation at the biotechnological laboratory of the University Cheikh Anta Diop of Dakar. The *Moringa oleifera* and *Pleurotus ostreatus* powders were combined in a 2:1 ratio to create the dietary supplement. The mixture was created following Kane *et al,* instructions (2017). The combination was dissolved in 0.01% starch paste before being fed to the rats.

b) *Animals and grouping*

Wistar rats strain to weigh 150 to 200 g were obtained from the Animal House of the National Institute of Youth and Sports in Yaounde. They were placed in plastic cages under standard laboratory conditions (temperature 20 to 30°C, relative air humidity 45 to 55%, and 12/12h light/dark cycle). The rats were fed with a basal diet and water *ad libitum*. The feed was a standard rat chow composed of carbohydrates (52%), protein (22%), fat (6.5%), water (12%), ash (6%), and fiber (4.5%). Every two days for 21 days, between 10:00 and 11:00 am, before the mixture administration of *Moringa oleifera* and *Pleurotus ostreatus* in proportion 2:1, made as reported by Kane *et al*., the amount of food and water ingested by each group of rats as well as body weights were recorded (2017). The experiments were performed during the day (09am–03pm).

c) *Experimental design*

Thirty (30) adult male and female Wistar rats weighing 150 to 200 g were separated into five groups of six after two weeks of acclimatization:

- Group 1 (TG): a stress-free control group that consumed only their regular diet of water, food, and vehicle (starch paste) once daily for 21 days,
- Group 2 (TP): a control group that received paracetamol 12 hours after ethanol administration, was supplied in five sequential doses of 2 g. kg-1 using an orogastric tube to stress the group. For 21 days, they consumed the standard diet of water and food at their leisure in addition to the vehicle starch paste,
- Group 3 (D1P): a group that received 500 mg/kg of FMP16 and was stressed by ethanol in five sequential doses of 2 g. kg^{-1}, administered through an orogastric tube; then received paracetamol 12 h after the last dose of ethanol. They received the standard diet (water and food ab-libitum) and the vehicle starch paste once a day for 21 days,
- Group 4 (D2P): a group that received 1000 mg/kg of FMP16 and was stressed by ethanol in five sequential doses of 2 g. kg^{-1}, administered through an orogastric tube; then received paracetamol 12 h after the last amount of ethanol. They received the normal diet (water and food ab-libitum) and the vehicle starch paste once a day for 21 days,
- Group 5 (D3P): a group that received 1500 mg/kg of FMP16 and was stressed by ethanol infive sequential doses of 2 g. kg^{-1}, administered through an orogastric tube; then received paracetamol 12 h after the last dose of ethanol. They received the standard diet (water and food ab-libitum) and the vehicle starch paste once a day for 21 days.The rats were given full access to food and water, and were on 12-hour light cycle each day (dark 12h-12h light). They were force-fed FMP16 using a gastroesophageal catheter and weighed every day. They fasted for the entire day before the animal sacrifice.

On the 23rd day, the rats were given a night of rest before being slaughtered (while sedated with ether) by having their jugular veins cut. Organs such the liver, kidneys, brain, and testicles were collected along with blood. The liver, which was exclusively used in this study, was wrung out, weighed, and stored at -20°C until

processing day. It was then rinsed with ice-cold saline (0.9% NaCl) to eliminate any remaining blood.

d) Determination of the biochemicals parameters in liver

- Preparation of liver supernates

Prior to biochemicals analysis, each liver sample was homogenized using a Poterproctor placed on ice and 10% homogenate was prepared using the KCL buffer solution (1.15%). The homogenates were centrifuged at 3000 rpm for 30 min at 4 ° C to collect the supernatant used for analysis. The supernatant of each sample was aliquoted in 1.5 ml Eppendorf tubes to estimatethe activity of antioxidant parameters (peroxidized lipids LPO, glutathione cellular GSH, catalase CAT). All liver parameters were expressed as activity per mg proteins. The proteins concentration in each fraction were determined by the method of (Gornall *et al*., 1949)

- Determination of biochemical parameters

- Using the method of thiobarbituric acid-reacting substances, the mean malondialdéhyde (MDA) level (mol/mg protein), a measure of lipid peroxidation, was evaluated (Singh *et al*., 2014).
- The level of catalase activity was assayed by the method of Sinha (1972).
- The level of Glutathionecellular activity was evaluated by the method of Ellman (1959).
- Serum glutamyl oxaloacétate transaminase (SGOT) and serum glutamyl pyruvate transaminase (SGPT) activities were assayed by the method of Karmen *et al*., (1955) and measured by standard essay kits SGM Italia Rome, Via Eschilo, 10139, (2012).
- The albumin level was assayed by the method of Ferreria& Price (1974) and measured by standard essay kits Hospitex diagnostics, Via Arno, 4001010L, (2013). Creatinin level was assayed by the method of Bergmeyer (1987) and measured by standard essay kits Hospitex diagnostics, Via Arno, 4001621L (2014).
- Testosterone level was assayed by the method of Tietz, (1986) and measured by Kit ELISA (DRG Diagnostics, Germany, EIA- 1559, (2009).
- Total Cholesterol level was assayed by the method of Allain *et al*, (1974) and measured by standard essay kits Hospitex Diagnostics, Via Arno, 4001210L, (2011).
- HDL Cholesterol level was assayed by the method of Grove (1979) and measured by standard essay kits SGM Italia, 10176, (2009).
- Triglycerides level was assayed by the method of Babblok *et al*., (1988) and measured by standard essay kits Fortress Diagnostics, United kingdom, BXC0271, (2013).

e) Statistical Analysis

IBM SPSS Statistics 20 software was used for statistical analysis and data processing. P-values less than 0.05 were regarded as significant in the statistical analysis, which was conducted using one-way analysis of variance (ANOVA) and Bonferroni's post-test for multiple comparisons. The results are presented as the mean and standard deviation (SD).

III. Results and Discussion

Results have shown that no significant difference was observed in final body weights (155–173g) (Table1). Body weight gain ranged between 0.8 and 19 g for the four treatment groups. A decrease of 10% in the weight of D3N group was observed. These results corroborate those of Alam *et al.,* (2011, 2009) who found that a diet enriched with *Pleurotus ostreatus* decreases the body weight of animals. However, Bobek *et al.,* (1998) have shown that it does not affectthe weight as well as Schneider *et al.,* (2011) who worked on humans with a daily dose of 30 g of dried oyster mushrooms, found that this does not affect anthropometric data. Bénissan *et al.,* 2012, showed that the daily intake of 30 g of *Moringa oleifera* leaves improves nutritional recovery in children suffering from malnutrition. Hanaa *et al.,* (2014), showed that a dose of 600 mg/kg of *Moringa oleifera* lowers the body mass index in obese subjects. Furthermore, the mixture of these species at a high dose of 1500 mg / kg, would explain the weight loss. This result was in contrast with those of Osman et al. (2012), who reported up to 14% changes in body weight of rats given *M.oleifera* extract for 21 days, attributing these changes to the rich nutrient quality of the extract.

Results also have shown no significant difference in the amount of protein in the liver (figure1). Regarding lipid peroxidation (figure 2), results show no significant difference in the concentration of peroxidized lipids between the groups except between the unstressed control group (TG) and the 1500 mg/kg dose group where the concentration was 34% higher. These results are not in agreement with those of Mladenovic *et al,* (2013); Patere *et al,* (2011); Johnsen *et al,* (2007). The effect of FMP16 on oxidative stress enzymes such as catalase and cellular glutathione was also studied. Our results showed an increase in catalase activity of 87%, 85%, 90%, 82% respectively for the TG, D1P, D2P and D3P groups compared to the TP group (intoxicated and untreated). Also, catalase activity of the 1000 mg/kg dose (D2P) was 35% and 43% higher respectively compared to the 500 and 1500 mg/kg doses (figure 3 and 4). These results corroborate those of Lamou et al, (2015); Pornariya and Kanok, (2009); Elmastas et al, (2007); Mishra et al, (2011) and, they would be justified by the antioxidant capacity of both *Moringa oleifera* and *Pleurotus ostreatus* (Zhang et al., 2016; Elmastas *et al.,* 2007; Makkar *et al.,* 1996; Sholapur and Patil, 2013).

The liver damage caused by paracetamol, known as a hepatotoxic agent in case of overdose

(Séide, 2008), is frequently used to assess the hepato-protective effects of medicinal plants (Lewerenz et al., 2003; Liu et al., 2011). The markers that are used to determine toxicity are usually transaminases (ALAT and ASAT), whose high concentration in the extracellular medium is synonymous with an alteration of the cells. In this study, our results showed that ALAT and ASAT activities were decreased in the FMP16 groups (Table 2). Thus, ALAT activity decreased by 29% in the 1000 mg/kg dose group compared to the intoxicated control. ASAT activity was higher in the intoxicated group (TP) by 28% and 26% compared to the 1000 and 1500 mg/kg doses. Compared to the results of the studies by Adedapo *et al,* (2009) and Alam *et al,* (2011), who instead found that a dose of 1600 mg/kg of *Moringa oleifera* leaves rather increased ALAT and ASAT activity; and on the other hand that a diet supplemented with 5% *Pleurotus* rather decreased transaminase activities, we could think that this explains the fact that FMP16 rather tends to regulate their activities due to the antagonistic effect that these two species have.

Furthermore, results showed that the administration of FMP16 did not cause any significant difference in albumin and testosterone levels (Table 2). These results are similar to those of Alam *et al,* (2011) and Adedapo *et al,* (2009) who found that administration of Pleurotus and *Moringa oleifera* had no effect on albumin levels. Howewer, Prabsattro et al, (2015), Zade et al, (2013), Okolo et al, 2016 rather found in their studies that *Moringa oleifera* increases sexual performance and thus could be considered as a potential aphrodisiac.

Regarding creatinine, FMP16 administration decreased creatinine levels in the treated groups (D1P, D2P, D3P) compared to the untreated and stressed group (TP) (Table 2). These results corroborate those of Sirag, (2009), Adedapo *et al,*(2009), Kane *et al,* 2022 who showed the protective effect of *Pleurotus ostreatus* and *Moringa oleifera* on kidney damage.

Our results on lipid metabolism in rats revealed a significant decrease in total cholesterol in the 500, 1000 and 1500 mg/kg dose groups (figure 5). There was a 28%, 39%, 30% and 38% difference in TP, D1P, D2P and D3P compared to TG. In addition, a difference of 15% and 14% of D1P and D3P compared to TP. However, there was no significant difference in HDL cholesterol levels (Table 2). The results of the Triglycerides levels (Table 2) show a difference of 47% and 41% of the 500 and 1000 mg/kg dose compared to the TG control group. There was also a 28% decrease in Triglycerides levels at the 500 mg/kg dose compared to the dose 1000 mg/kg. In most of the studies on the effects of *Pleurotus ostreatus* and *Moringa oleifera,* they found a decrease in the concentration of LDL cholesterol which is more related to cardiovascular diseases (Bobek and Galbavy, 1999; Bobek *et al.,* 1998; Hossain *et al.,* 2003). These results are also in agreement with those of Alam *et al,* (2009), Schneider et al, (2011), Chumark *et al,* (2008), Kane *et al,* (2022). Our results (figure 6) and those of previous studies suggest that FMP16 would be an excellent cholesterol-lowering agent that could be recommended for the prevention and treatment of cardiovascular diseases.

Table 1: Effect of the dietary supplement on rat weights

GROUPES	Starting Body weight (g)	Final Body weight (g)	P-value*
TG	154 ± 3,34	173,67 ± 9,16 [a]	0, 02
TP	154,33±3,44	177,33±4,84 [d]	0,01
D1P	154±2,53	185,67±2,86 [e]	0,01
D2P	153,20±2,68	168±4,14	0,6
D3P	153±3,03	157±7,14	0, 13

**ANOVA test; TG: control group; TP: stressed group ethanol+paracetamol; D1P: stressed and treated group 500 mg/kg: D2P: stressed and treated group 1000 mg/kg; D3P: stressed and treated group 1500 mg/kg; a, d, e: mean statistically different with D3P à p <0,05 (test de Bonferoni)*

Table 2: Effects of dietary supplement FMP16 on serum transaminases, albumin, testosterone, HDL-cholesterol and triglycerides activity

GROUPS	ALAT (U/l)	ASAT (U/l)	ALBUMINE (g/dl)	TESTOS. (ng/dl)	HDL-C (mg/dl)	Triglyc. (mg/dl)
TG	32,74±7,09	160,92±30,02	1,53±0,20	0,31±0,06	39,07±2,18	52,03±0,68
TP	48,48±3,32	196,35±33,40	1,61±0,37	0,47±0,12	38,88±1,88	78,14±12,69
D1P	43,16±8,80	209,16±25,74	1,80±0,10	0,53±0,08	36,30±2,89	71,02±18,92
D2P	34,31±4,55	141,86±11,19	1,60±0,20	0,44±0,04	35,08±2,53	98,18±7,64
D3P	43,66±2,13	145,86±22,20	1,75±0,18	0,43±0,03	34,17±5,47	87,83±21,97

The values are expressed as mean ± SD. TG: Control group rats with food and water ad libitum, TP: stressed rats without treatment, D1P: dose of 500 mg/kg, D2P : dose of 1000 mg/kg, D3P: dose of 1500 mg/kg.

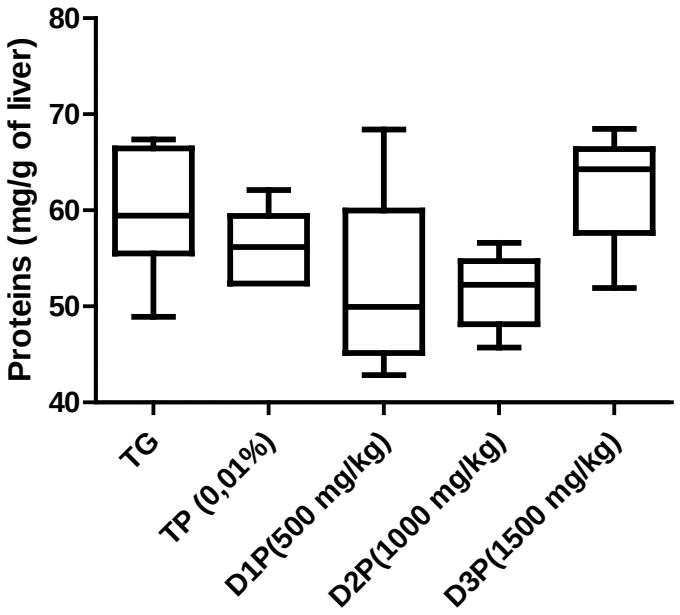

Figure 1: Total protein concentrations (mg/g of liver) in groups stressed by swimming and the Ethanol+ paracetamol combination

The values are expressed as mean ± SD. TG: Control group rats with food and water ad libitum, TP: stressed rats without treatment, D1P: dose of 500 mg/kg, D2P: dose of 1000 mg/kg, D3P: dose of 1500 mg/kg.

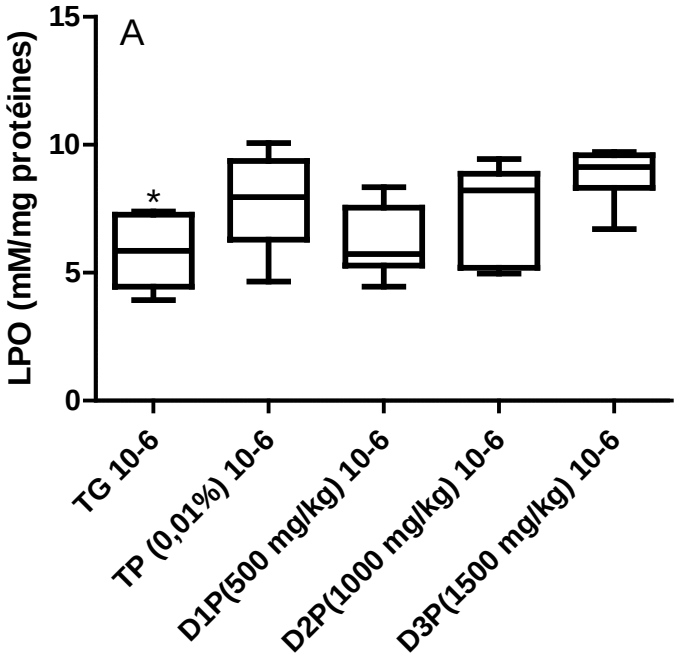

Figure 2: Effect of the dietary supplement on concentrations of peroxidized lipids (LPO)

The values are expressed as mean ± SD. TG: Control group rats with food and water ad libitum, TP: stressed rats without treatment, D1P: dose of 500 mg/kg, D2P: dose of 1000 mg/kg, D3P : dose of 1500 mg/kg. 10-6

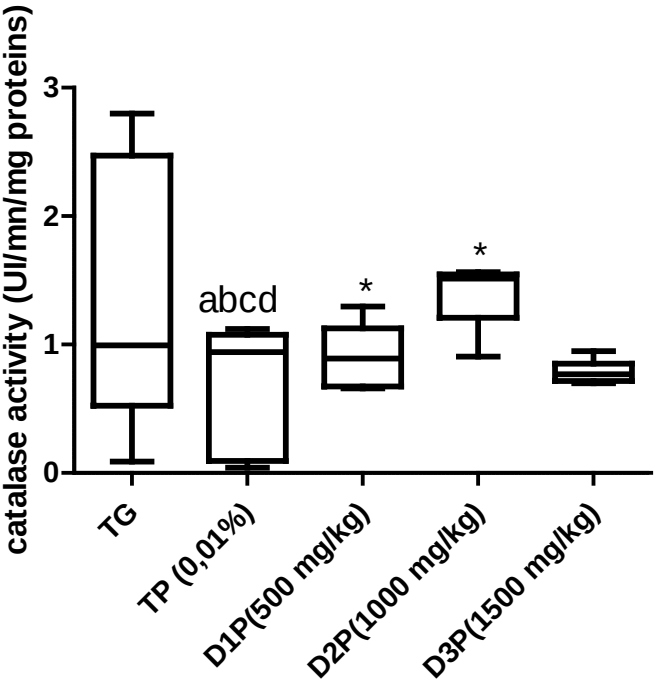

Figure 3: Effect of the dietary supplement on catalase activity (UI/mn/mg proteins)

The values are expressed as mean ± SD. TG: Control group rats with food and water ad libitum, TP: stressed rats without treatment, D1P : dose of 500 mg/kg, D2P : dose of 1000 mg/kg, D3P: dose of 1500 mg/kg.
a, b, c, d, mean statistically different with TG, D1P, D2P et D3P à p<0,05 (Test de Bonferroni)
**, +, mean statistically different with D2P and D3P à p<0, 05 (Test de Bonferroni)*

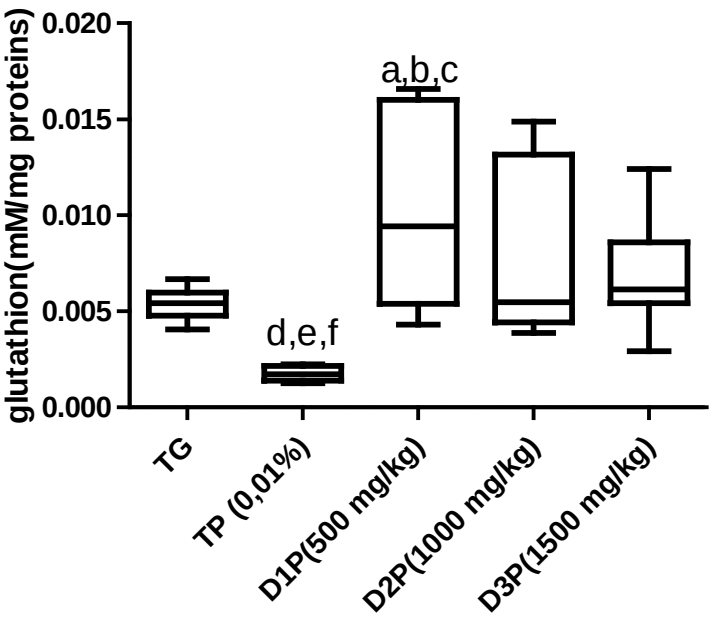

Figure 4: Effect of the dietary supplement on glutathion cellular activity (mM/mg proteins)
Values are means ± SD.

a , b, c, d mean statistically different with TG, D1P, D2P et D3P à p<0,05 (Test de Bonferroni)
e, f, g mean statistically different with TG, D2P et D3P à p<0,05 (Test de Bonferroni)

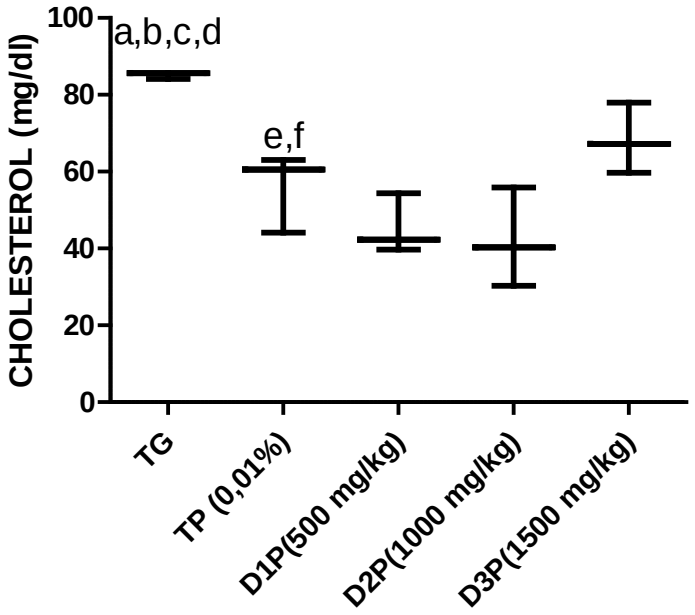

Figure 5: Total cholesterol level (mg/dl) in serum of rats stressed by the combination of ethanol (30% - 2g/kg) and paracetamol (750mg/kg)

The values are expressed as mean ± SD. TG: Control group rats with food and water ad libitum, TP: stressed rats without treatment, D1P: dose of 500 mg/kg, D2P: dose of 1000 mg/kg, D3P: dose of 1500 mg/kg.
a, b ,c, d statistically different mean compared to TP, D1P, D2P and D3P at $p<0.05$ (LSD test)
e, f mean statistically different from D1P and D3P at $p<0.05$ (LSD test)

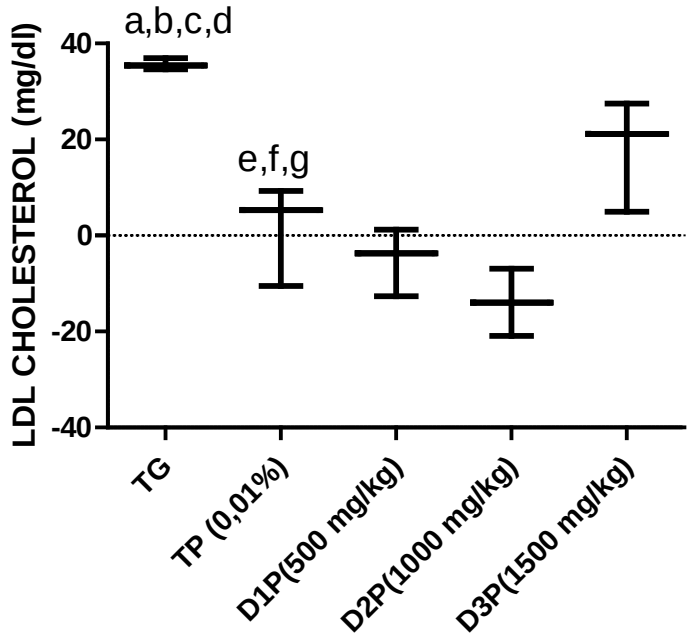

Figure 6: LDL cholesterol levels (mg/dl) in serum of rats stressed by the combination of ethanol (30% - 2g/kg) and paracetamol (750mg/kg)

The values are expressed as mean ± SD. TG: Control group rats with food and water ad libitum, TP: stressed rats without treatment, D1P: dose of 500 mg/kg, D2P: dose of 1000 mg/kg, D3P: dose of 1500 mg/kg.
a,b,c ,d statistically different mean compared to TP, D1P, D2P and D3P at $p<0.05$ (LSD test)
e, f, g mean statistically different from D1P, D2P and D3P at $p<0.05$ (LSD test)

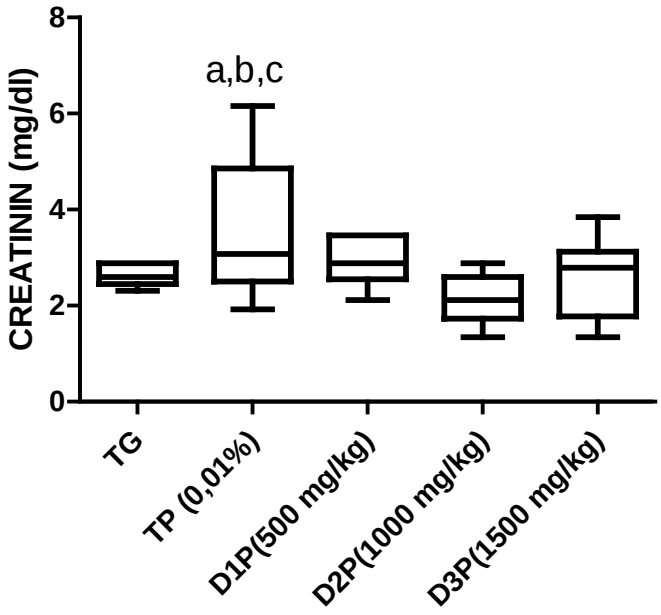

Figure 7: Effect of the dietary supplement on creatinin activity (mg/dl) in the serum of rats stressed by the combination of ethanol (30% - 2g/kg) and paracetamol (750mg/kg)

The values are expressed as mean ± SD. TG: Control group rats with food and water ad libitum, TP: stressed rats without treatment, D1P: dose of 500 mg/kg, D2P: dose of 1000 mg/kg, D3P: dose of 1500 mg/kg.a, b mean statistically different from D2P and D3P at $p<0.05$ (Bonferroni test)

IV. Conclusion

A dietary supplement of Moringa oleifera leaves and Pleurotus ostreatus in wistar rats shows that the powders of M. oleifera leaves and P. ostreatus mixture have an antihyperlipidemic effect as it significantly lowers total and LDL cholesterol levels in rats stressed by combination of ethanol and paracetamol. The dose 1000 mg/kg is most appropriate for chemically stressed animals. FMP16 would have no effect on albumin and testosterone levels.

Acknowledgements

The authors would like to thank the Biotechnology Laboratory of the University Cheikh Anta Diop of Dakar, Senegal, and the Faculty of Science, the FODRUS-LAPHER-Biotech of the University of Yaoundé I, Cameroon. The project was part of a scholarship provided to Fatou Corka KANE through Africa for Innovations, Mobility, Exchange, Globalization and Quality (AFIMEGQ) program sponsored by the European Commission's EACEA program.

Conflict of Interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

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Global Journal of Medical Research: L
Nutrition & Food Science
Volume 22 Issue 2 Version 1.0 Year 2022
Type: Double Blind Peer Reviewed International Research Journal
Publisher: Global Journals Inc. (USA)
Online ISSN: 2249-4618 & Print ISSN: 0975-5888

Comparison of Taste and Smell Test Results Before and After COVID-19 in Yakumo Residents Health Checkup Comparison between 2019 and 2022

By Naomi Katayama
Nagoya Womens University

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Taste and smell tests were conducted on the study participants, and the results have been reported.

However, in 2020 and 2021, we were could not receive a health checkup data to the influence of the new coronavirus.

But, in August 2022, we were finally able to obtain the results of taste and smell tests.

Therefore, in this study, we compare the taste and smell test results obtained in August 2019 (before the COVID-19 epidemic) and in August 2022 (after the COVID-19 epidemic).

Taste and smell were measured using a simple test kit, and height, weight, and blood pressure were also obtained.

Keywords: simple salty taste test, simple olfactory test, resident medical examination, age group.

GJMR-L Classification: DDC Code: 004.7 LCC Code: QA76.889

Strictly as per the compliance and regulations of:

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Taste and smell were measured using a simple test kit, and height, weight, and blood pressure were also obtained.

129 males and 169 females participated in 2019 and 142 males and 202 females participated in 2022.

Each result was compared by age group (the 40s, 50s, 60s, 70s, 80s).

As our results, there were no statistically significant differences in gender, age, height, weight, systolic blood pressure, diastolic blood pressure, and salty taste test results in each age group between 2019 and 2022 (unpaired student t-test or Mann-Whitney test).

However, the olfactory test results were statistically significantly lower in 2022 than in 2019.

Keywords: simple salty taste test, simple olfactory test, resident medical examination, age group.

I. Introduction

From 2007 to 2019, every August in Yakumo Town, Hokkaido, the authors examined the sense of taste and olfactory tests during a health checkup for residents[1-12].

However, in 2020 and 2021, we could not undergo a medical examination due to the COVID-19 epidemic.

As the COVID-19 epidemic has subsided, this fiscal year (August 2022), Hokkaido, August.

We obtained the taste and smell test results during the health checkup for the residents of Yakumo Town.

Therefore, we compared the taste and smell test results obtained in 2019 and the taste and smell test results obtained in 2022.

I decided to confirm whether or not there was an impact of COVID-19 by comparing two data.

II. Material and Method

Among the participants in the health checkup for Yakumo town residents were measured for height, weight, blood pressure (systolic and diastolic), salty taste tests, and olfactory tests.

There were 298 subjects (129 males 169 females) in 2019.

And there were 344 subjects (142 males, 202 females) in 2022.

Survey items comparing 2019 and 2022 are age, height, weight, systolic blood pressure, diastolic blood pressure, the results of a simple olfactory test, and the results of a simple salty taste test.

The results of the simple salty taste test were performed by using Salsive (manufactured by Advantech). The Salsive is the filter paper. Salsive comes in 6 different salt concentrations (0.6% 0.8%, 1.0% 1.2%, 1.4%, 1.6%). Participants put Salsive in their mouth to check the salty taste.

Concentration was recorded when participants perceived salty teste[13].

The results of the simple olfactory test were performed using an odor stick (Daiichi Yakuhin Kogyo Co., Ltd.).

Twelve kinds of odors are applied to the filter paper (Japanese ink, wood, perfume, menthol, mandarin orange, curry, household gas, roses, cypress, stuffy socks/sweaty, condensed milk, fried garlic). The number of odors perceived by participants was recorded.

The obtained data were statistically processed by sex and age groups.

2019 and 2022 data were F-tested, and the results were either unpaired Student-t test or Mann.

Whitney test was performed to confirm the presence or absence of statistical significance.

a) Ethical review board

This study conducted with the approval of the Ethical Review Board (Nagoya women's University Ethics Committee: "hitowomochiitakennkyuuni-kannsuruiinnkai"). The approval number is 2019-26.

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III. Result

There were 298 participants (129 male and 169 female) in 2019, and 344 participants (142 male and 202 female) in 2022. The distribution of each age group is shown in Table 1. In both years, there were many participants in their 60s and 70s.

Table 1. Age composition of participants in 2019 and 2022 (number of people)

	40s	50s	60s	70s	80s	Total
2019 Male	10	24	49	40	6	129
2019 Female	23	40	66	37	3	169
2022 Male	13	20	38	59	12	142
2022 Female	34	37	64	57	10	202

Table 2 shows the average values and standard deviations by age group for each inspection item in FY2019.

The average systolic blood pressure for both males and females in their 70s and 80s was 140 mmHg, exceeding the normal range.

However, the average diastolic blood pressure was 90 mmHg or less in both men and women, which was within the normal range.

The average value of the simple olfactory test results in the 80s female was six, and half of the twelve types of odors could be recognized. All females of other ages had a simple olfactory test result of six or higher.

However, the average value for males was six or less, resulting in a less recognizable odor.

The average value of salty taste test results for women in their 80s exceeds hers by 1.0%.

But otherwise, both males and females, in the age-specific salty taste test results, salty taste could be recognized less than 1.0%.

Table 2. FY2019 Yakumo Town Resident Health Check Basice Data (169 Female, 129 Male)

Female	40s		50s		60s		70s		80s	
	Average	S D	Average	S D	Average	S D	Average	S D	Average	S D
Age	45.22	2.61	54.33	3.04	64.52	2.77	72.84	2.57	82.00	2.00
Hight	158.01	5.17	155.52	6.01	153.80	5.15	150.56	5.38	147.37	2.84
Weight	57.15	11.48	56.42	9.08	55.66	8.91	52.82	10.01	49.57	11.37
Systolic blood pressure	122.26	15.75	131.58	20.57	137.14	19.05	140.11	24.48	149.00	29.44
Diastolic blood pressure	70.13	10.11	77.35	12.95	77.05	11.94	74.70	11.33	77.00	7.00
Olfactory test results	9.26	1.91	9.60	1.81	8.94	2.37	7.43	2.22	6.33	2.08
Salty taste test results	0.88	0.37	0.87	0.37	0.85	0.35	0.90	0.39	1.07	0.64

	40s		50s		60s		70s		80s	
Male	Average	S D	Average	S D	Average	S D	Average	S D	Average	S D
Age	45.50	3.21	54.83	3.14	64.84	3.32	73.03	3.17	84.83	3.76
Hight	170.05	4.63	167.96	6.29	167.28	5.89	164.69	5.35	159.13	1.75
Weight	74.15	11.32	71.34	8.93	68.93	9.35	66.23	10.08	63.50	6.39
Systolic blood pressure	136.80	18.35	131.00	18.98	138.27	14.50	145.53	24.70	134.67	14.94
Diastolic blood pressure	80.90	14.36	81.33	11.34	83.12	8.70	79.73	15.39	66.17	9.02
Olfactory test results	9.00	2.00	8.13	2.15	7.18	2.34	6.49	3.27	5.67	2.80
Salty taste test results	0.90	0.33	0.92	0.47	0.89	0.38	0.94	0.46	0.90	0.21

Table 3 shows the average values and standard deviations by age group for each inspection item in FY2022.

In females, the average systolic blood pressure in their 70s and 80s is over 140 mmHg, which exceeds the normal range.

And also in males, the average systolic blood pressure in their 80s is over 140 mmHg, which exceeds the normal range.

However, the mean diastolic blood pressure for both males and females was below 90 mmHg, which was within the normal range.

Females in their 80's and males in their 80's and 70's recognized six or less of the twelve odors. As a result, olfactory recognition decreased with age.

The results of the salty taste test showed that they could recognize less than 1.0% salty taste for both males and females.

Table 3. FY2022 Yakumo Town Resident Health Check Basice Data (202 Female, 142Male)

Female	40s		50s		60s		70s		80s	
	Average	S D	Average	S D	Average	S D	Average	S D	Average	S D
Age	44.85	2.65	55.08	2.95	65.02	3.00	73.84	2.77	82.50	2.46
Hight	156.21	10.80	157.30	5.44	174.61	169.53	151.48	6.46	149.02	6.68
Weight	55.26	11.65	68.28	74.94	54.65	10.21	59.21	36.86	53.38	12.22
Systolic blood pressure	122.59	22.51	131.95	20.20	135.20	19.31	144.80	20.63	149.70	16.81
Diastolic blood pressure	70.18	11.45	75.03	14.33	76.30	11.91	77.11	12.83	75.30	11.96
Olfactory test results	8.44	2.70	8.78	2.11	8.66	2.54	6.16	2.65	5.90	2.47
Salty taste test results	0.88	0.33	0.72	0.19	0.81	0.31	0.85	0.31	0.64	0.08

	40s		50s		60s		70s		80s	
Male	Average	S D	Average	S D	Average	S D	Average	S D	Average	S D
Age	46.00	3.14	53.90	2.75	63.66	2.68	73.63	2.41	84.67	3.89
Hight	168.51	7.80	168.45	5.49	167.91	6.13	164.59	5.88	159.70	7.10
Weight	78.64	19.32	71.61	10.67	70.14	8.93	65.49	9.75	63.61	10.38
Systolic blood pressure	131.15	16.12	130.85	16.79	135.61	18.27	137.32	21.84	144.92	20.75
Diastolic blood pressure	77.8	17.2	79.9	10.6	79.8	9.2	76.7	12.9	72.9	13.8
Olfactory test results	6.38	2.53	8.15	2.43	6.61	3.03	5.72	3.06	3.58	2.87
Salty taste test results	0.89	0.41	0.81	0.28	0.91	0.36	0.89	0.41	0.97	0.46

The results of 2022 and 2019 were compared using statistical methods.

The results of comparing the age distribution of females in 2022 and 2019 showed Table 4. As a result, there was no statistically significant difference between 2022 and 2019.

Table 4 Age Comparison Results for 2019 and 2022 Participants Female (169 in 2019, 202 in 2022)

	40s		50s		60s	
	2019	2022	2019	2022	2019	2022
F-test	P=0.476		P=0.422		P=0.260	
unpaired-t test	P=0.611		p=0.272		p=0.326	
Mann-Whitney test						
	70s		80s		Total	
	2019	2022	2019	2022	2019	2022
F-test	P=0.295		P=0.405		p=0.022	
unpaired-t test	P=0.086		p=0.756		p=0.134	
Mann-Whitney test						

The results of comparing the age distribution of males in 2022 and 2019 showed Table 5. As a result, there was no statistically significant difference between 2022 and 2019.

Table 5 Age Comparison Results for 2019 and 2022 Participants Male (129 in 2019, 142 in 2022)

	40s		50s		60s	
	2019	2022	2019	2022	2019	2022
F-test	P=0.481		P=0.264		P=0.081	
unpaired-t test	P=0.199		p=0.306		p=0.082	
Mann-Whitney test						
	70s		80s		Total	
	2019	2022	2019	2022	2019	2022
F-test	P=0.039*		P=0.293		p=0.119	
unpaired-t test			p=0.662		p=0.199	
Mann-Whitney test	p=0.063					

The results of comparing the height distribution of females in 2022 and 2019 showed Table 6. As a result, there was no statistically significant difference between 2022 and 2019.

Table 6 Hight Comparison Results for 2019 and 2022 Participants Female (169 in 2019, 202 in 2022)

	40s		50s		60s	
	2019	2022	2019	2022	2019	2022
F-test	P=0.0001**		P=0.263		P=0.443	
unpaired-t test			P=0.177		P=0.653	
Mann-Whitney test	P=0.987					
	70s		80s		Total	
	2019	2022	2019	2022	2019	2022
F-test	P=0.210		P=0.093		P=0.003**	
unpaired-t test	P=0.626		P=0.631			
Mann-Whitney test					P=0.311	

The results of comparing the height distribution of males in 2022 and 2019 showed Table 7. As a result, there was no statistically significant difference between 2022 and 2019.

Table 7 Hight Comparison Results for 2019 and 2022 Participants Male (129 in 2019, 142 in 2022)

	40s		50s		60s	
	2019	2022	2019	2022	2019	2022
F-test	P=0.063		P=0.262		P=0.392	
unpaired-t test	P=0.586		P=0.786		P=0.631	
Mann-Whitney test						
	70s		80s		Total	
	2019	2022	2019	2022	2019	2022
F-test	P=0.248		P=0.001**		P=0.115	
unpaired-t test	P=0.960				P=0.575	
Mann-Whitney test			P=0.235			

The results of comparing the weight distribution of females in 2022 and 2019 showed Table 8. As a result, there was no statistically significant difference between 2022 and 2019.

Table 8 Weight Comparison Results for 2019 and 2022 Participants Female (169 in 2019, 202 in 2022)

	40s		50s		60s	
	2019	2022	2019	2022	2019	2022
F-test	P=0.480		P=0.317		P=0.138	
unpaired-t test	P=0.547		P=0.819		P=0.550	
Mann-Whitney test						
	70s		80s		Total	
	2019	2022	2019	2022	2019	2022
F-test	P=0.450		P=0.510		P=0.146	
unpaired-t test	P=0.668		P=0.641		P=0.548	
Mann-Whitney test						

The results of comparing the weight distribution of males in 2022 and 2019 showed Table 9. As a result, there was no statistically significant difference between 2022 and 2019.

Table 9 Weight Comparison Results for 2019 and 2022 Participants Male (129 in 2019, 142 in 2022)

	40s		50s		60s	
	2019	2022	2019	2022	2019	2022
F-test	P=0.049*		P=0.201		P=0.377	
unpaired-t test			P=0.928		P=0.544	
Mann-Whitney test	P=0.789					
	70s		80s		Total	
	2019	2022	2019	2022	2019	2022
F-test	P=0.414		P=0.130		P=0.033**	
unpaired-t test	P=0.781		P=0.876			
Mann-Whitney test					P=0.776	

The results of comparing the systolic blood pressure distribution of females in 2022 and 2019 showed Table 10. As a result, there was no statistically significant difference between 2022 and 2019.

Table 10 Systolic Blood Presure Comparison Results for 2019 and 2022 Participants Female (169 in 2019, 202 in 2022)

	40s		50s		60s	
	2019	2022	2019	2022	2019	2022
F-test	P=0.038*		P=0.453		P=0.455	
unpaired-t test			P=0.937		P=0.567	
Mann-Whitney test	P=0.552					
	70s		80s		Total	
	2019	2022	2019	2022	2019	2022
F-test	P=0.147		P=0.193		P=0.363	
unpaired-t test	P=0.343		P=0.958		P=0.618	
Mann-Whitney test						

The results of comparing the systolic blood pressure distribution of males in 2022 and 2019 showed Table 11. As a result, there was no statistically significant difference between 2022 and 2019.

Table 11 Systolic Blood Presure Comparison Results for 2019 and 2022 Participants Male (129 in 2019, 142 in 2022)

	40s		50s		60s	
	2019	2022	2019	2022	2019	2022
F-test	P=0.346		P=0.281		P=0.064	
unpaired-t test	P=0.442		P=0.978		P=0.451	
Mann-Whitney test						
	70s		80s		Total	
	2019	2022	2019	2022	2019	2022
F-test	P=0.185		P=0.216		P=0.495	
unpaired-t test	P=0.117		P=0.300		P=0.292	
Mann-Whitney test						

The results of comparing the diastolic blood pressure distribution of females in 2022 and 2019 showed Table 12. As a result, there was no statistically significant difference between 2022 and 2019.

Table 12 Diastolic Blood Pressure Comparison Results for 2019 and 2022 Participants Female (169 in 2019, 202 in 2022)

	40s		50s		60s	
	2019	2022	2019	2022	2019	2022
F-test	P=0.269		P=0.266		P=0.493	
unpaired-t test	P=0.988		P=0.460		P=0.721	
Mann-Whitney test						
	70s		80s		Total	
	2019	2022	2019	2022	2019	2022
F-test	P=0.207		P=0.206		P=0.913	
unpaired-t test	P=0.324		P=0.822		P=0.747	
Mann-Whitney test						

Table 13 shows the results of comparing males' systolic blood pressure by age group.

Although there was no statistically significant difference by age group, P<0.05 (P=0.045*) for all age groups.

The results showed that the diastolic blood pressure in 2022 was statistically significantly lower than the diastolic blood pressure in 2019.

Table 13 Diastolic Blood Pressure Comparison Results for 2019 and 2022 Participants Male (129 in 2019, 142 in 2022)

	40s		50s		60s	
	2019	2022	2019	2022	2019	2022
F-test	P=0.286		P=0.343		P=0.346	
unpaired-t test	P=0.648		P=0.669		P=0.090	
Mann-Whitney test						
	70s		80s		Total	
	2019	2022	2019	2022	2019	2022
F-test	P=0.119		P=0.172		P=0.438	
unpaired-t test	P=0.327		P=0.312		P=0.045*	
Mann-Whitney test						

Table 14 shows the results of a comparison of females' olfactory test results by age group.

A statistically significant difference comes out in their seventies. In 2022, olfactory recognition was statistically significantly lower than in 2019 (P<0.05: P=0.024*). Comparing the results of the olfactory cognition test in 2022 and 2019, there was no statistically significant difference in each age group. However, as a result of the overall comparison, olfactory recognition was statistically significantly lower (P<0.01: P=0.001**) in 2022 than in 2019.

表14 Olfactory test results Comparison Results for 2019 and 2022 Participants Female (169 in 2019, 202 in 2022)

	40s		50s		60s	
	2019	2022	2019	2022	2019	2022
F-test	P=0.044*		P=0.170		P=0.284	
unpaired-t test			P=0.072		P=0.512	
Mann-Whitney test	P=0.257					
	70s		80s		Total	
	2019	2022	2019	2022	2019	2022
F-test	P=0.130		P=0.432		P=0.006**	
unpaired-t test	P=0.024*		P=0.789			
Mann-Whitney test					P=0.001**	

Table 15 shows the results of a comparison of male olfactory test results by age group.

A statistically significant difference comes out when he is in the 40s. In 2022, olfactory recognition was statistically significantly lower than in 2019 (P<0.05: P=0.014*).

Comparing the results of the olfactory cognition test in 2022 and 2019, other were no statistically significant difference in each age group. However, as a result of the overall comparison, olfactory recognition was statistically significantly lower (P<0.01: P=0.005**) in 2022 than in 2019.

Table 15 Olfactory test results Comparison Results for 2019 and 2022 Participants Male (129 in 2019, 142 in 2022)

	40s		50s		60s	
	2019	2022	2019	2022	2019	2022
F-test	P=0.229		P=0.281		P=0.049*	
unpaired-t test	P=0.014*		P=0.971			
Mann-Whitney test					P=0.568	
	70s		80s		Total	
	2019	2022	2019	2022	2019	2022
F-test	P=0.282		P=0.516		P=0.095	
unpaired-t test	P=0.315		P=0.138		P=0.005*	
Mann-Whitney test						

Table 16 shows the results of comparing females' salt taste tests by age group. Comparing the results of the salt taste cognition test in 2022 and 2019, other were no statistically significant difference in each age group.

Table 16 Salty taste test results Comparison Results for 2019 and 2022 Participants Female (169 in 2019, 202 in 2022)

	40s		50s		60s	
	2019	2022	2019	2022	2019	2022
F-test	P=0.305		P=0.001**		P=0.144	
unpaired-t test	P=0.985				P=0.501	
Mann-Whitney test			P=0.087			
	70s		80s		Total	
	2019	2022	2019	2022	2019	2022
F-test	P=0.060		P=0.003**		p=0.001*	
unpaired-t test	P=0.482					
Mann-Whitney test			P=0.093		P=0.187	

Table 17 shows the results of comparing males' salt taste tests by age group. Comparing the results of the salt taste cognition test in 2022 and 2019, other were no statistically significant difference in each age group.

Table 17 Salty taste test results Comparison Results for 2019 and 2022 Participants Male (129 in 2019, 142 in 2022)

	40s		50s		60s	
	2019	2022	2019	2022	2019	2022
F-test	P=0.261		P=0.019*		P=0.342	
unpaired-t test	P=0.962				P=0.807	
Mann-Whitney test			P=0.365			
	70s		80s		Total	
	2019	2022	2019	2022	2019	2022
F-test	P=0.255		P=0.005*		P=0.265	
unpaired-t test	P=0.597				P=0.551	
Mann-Whitney test			P=0.585			

IV. Discussion

For both male and female participants, age, height, and weight were not statistically significantly differences for comparison between 2019 and 2022. Females had no statistically significant difference in blood pressure between 2019 and 2022. However, there was no significant difference in diastolic blood pressure among males by age group, but when compared overall, the year 2022 was lower than in 2019. There was no statistically significant difference in cognition between 2019 and 2022 for salty taste. Regarding the sense of smell, there will be a statistically significant ($P<0.05$) decline in cognition in 2022 compared to 2019.

Whether this is due to the COVID-19 epidemic cannot be determined based on the results of this test alone. However, the results of this olfactory cognition test showed that the olfactory cognition in 2022 was lower than the olfactory cognition in 2019.

Therefore, we believe that it is necessary to continue to investigate the participants' sense of smell. At that time, we think it is needed to investigate COVID-19 morbidity as well. We believe it is necessary to track individuals individually.

Previous studies have reported a positive correlation between salt intake and blood pressure[15-19]. Therefore, in Japan and overseas, guidance to reduce salt intake is being carried out. Future studies will investigate the relationship dietary habits and blood pressure. It is necessary to investigate this in more detail. Relations with aging[20] and Alzheimer's disease[21,22] have also been reported regarding the decline in olfactory cognition. We could like to continue research on regional differences in Japan and clarify the results.

V. Conclusion

We compared taste and smelled simple test results before COVID-19 (2019) and after COVID-19 (2022). As a result, no statistically significant difference was observed in preference in all ages between 2019 and 2022. However, 2022 tended to have fewerol factory perceptions in all ages than in 2019.Butthe smell was a statistically significant difference between 2019 and 2019 in the total participants. Compared to 2022, the value tends to be lower in 2022, with a significant difference overall, and 2022 is not recognizable. It was found that the number of certain odors decreased in 2022. However, on this data, it cannot be concluded that the decline in olfactory recognition in 2022 was due to COVID-19.

In the future, we would like to clarify the presence or absence of regional differences by conducting surveys on more items and comparing them.

Acknowledgments

This research was partially supported by the research aid of Choju-iryo-kenkyu-kaihatsuhi, 2022 (30-14, Hirokazu Suzuki) and Japanese Society of Taste Technology, 2021 (Naomi Katayam) and the Ministry of Education, Science, Sports and Culture, Grant-in Aid for Scientific Research(C), 2020-2022 (20K02372, Naomi Katayam). We would like to express my deepest gratitude here.

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Global Journal of Medical Research: L
Nutrition & Food Science
Volume 22 Issue 2 Version 1.0 Year 2022
Type: Double Blind Peer Reviewed International Research Journal
Publisher: Global Journals Inc. (USA)
Online ISSN: 2249-4618 & Print ISSN: 0975-5888

Result of a Questionnaire Survey of 61 Females on their Satisfaction with Fish, Calcium and Protein Intake

By Naomi Katayama
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Abstract- Since the calcium intake has not reached the target amount of 600 ~ 800 mg per day in the Japanese diet, calcium intake is recommended in the daily diet. And in recent years, Japan has been promoting protein intake for the elderly to prevent sarcopenia and frailty. Japan also recommended to take EPA and DHA to maintain smooth blood vessels and memory maintenance. Therefore, as a first step to understanding the actual situation, this study conducted a questionnaire survey on the information of fish rich in EPA and DHA and the intake of Ca and protein.

Sixty-one participants were given a self-anubnustered questionnaire regarding fish, calcium and protein intake. The questionnaire method is self-administered, and the questions are about age, height, weight, and desired purchase price of fish. We asked the following questions, frequency of eating fish, frequency of eating meat, frequency of eating eggs, and frequency of eating bean products.

Keywords: questionnaire surveys, consumption of fish, calcium intake, protein intake.

GJMR-L Classification: DDC Code: 572.511 LCC Code: QP535.C2

Strictly as per the compliance and regulations of:

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Keywords: questionnaire surveys, consumption of fish, calcium intake, protein intake.

I. Introduction

In Japan, the Ministry of Health, Labor and Welfare publishes dietary standards for Japanese every five years. According to the Japanese Dietary Intake Standards issued by the Ministry of Health, Labor and Welfare in 2020, the daily protein intake for adults is 60g for males and 50g for females.

Per capita consumption of seafood in Japan continues to decline. According to the Japanese "Food Supply and Demand Chart," the consumption of edible seafood per person per year, peak intake of fish per person is 40.2 kg in 2001, but peak intake of fish per person is 24.6 kg in 2016 which is 1.1 kg less than the previous year. This fish intake is about the same level as in the late 1930s.In recent years, in Japan, protein intake is starting to decrease. This is thought to be due to factors such as the aging of the population. Calcium intake in Japan is much less than the recommended amount of 800～700mg for males and 650～600mg for females. The recent calcium intake in Japan is 470～550mg for males and 400～500mg for females. That is nearly 250mg less than the recommended amount.

Data from 1995 to 2015 show that Japanese protein intake is declining. Total protein may be sufficient, but animal proteins are too few, and it can be said that the Japanese lack “good quality protein.”That means that the amino acid score does not exceed 100.

The 61 females (18-28) who participated in a health class were asked about satisfaction with their intake of fish, calcium, and protein intake.

II. Materials and Methods

Participants were 61 females who were briefed about the study and signed a consent form. The questions consisted of the following seven items. 1) Do you like fish to eat? 2) Do you know a commercial fish that can eat up to the bones? 3) Would you like to eat a

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fish that can be eaten up to the bones of a commercial product? 4) Do you feel that you are deficient in calcium intake daily? 5) Do you want to take calcium positively? 6) Do you feel that you are deficient in protein intake daily? 7) Do you want to take protein positively? Participants self-administered responses to a seven-item questionnaire. In addition, participants were asked about their frequency of consumption of fish, meat, eggs, and beans using a self-administered questionnaire. The participants also answered the desired purchase price of the fish whose bones are edible.

III. Results

61 participants aged 18 to 28 years (see Table 1.), and the average age ± standard deviation was 21.1 ± 2.0. Weight was 52.9±7.1, and height was 158.4±5.9 (see Table 2.).

Table 1. Age distribution of 61 participants

years old	18	19	20	21	22	23	24	25	26	27	28
number of participants	3	7	14	20	7	2	3	2	2	0	1

Table 2. Basic information of 61 participants

	Average age	Average height	Average weight
Average±Standard diviation	21.1 ±2.0	158.4 ±5.9	52.9 ±7.1

Table 3 shows the results of a seven-item self-administered questionnaire conducted on the participants. Among the participants, 83.6% answered that they like to fish. The participants of 75.4% responded that they know a commercial fish that can eat up to the bones. Among the participants, 82.0% answered that they would like to eat a commercial product of fish whose bones are edible. In addition, 68.9% of the participants answered that they felt calcium deficiency in their daily lives. And 95.1% of the participants insisted on positive calcium intake. Similarly, 32.8% of the participants felt that they were not getting enough protein in their daily lives. The participants of 90.2% responded that they want to take protein positively.

Table 3. Questionnaire survery results on fish, calcium and protein intake of 61 participants (%)

Question items	Yes	No	No answer
Do you like fish to eat?	83.6	16.4	0.0
Do you know a commercial fish that can eat up to the bones?	75.4	24.6	0.0
Would you like to eat a fish that can be eaten up to the bones of a commercial product?	82.0	18.0	0.0
Do you feel that you are deficient in calcium intake on a daily basis?	68.9	29.5	1.6
Do you want to take calcium positively?	95.1	3.3	1.6
Do you feel that you are deficient in protein intake on a daily basis?	32.8	67.2	0.0
Do you want to take protein positively?	90.2	9.8	0.0

Participants filled out a self-administered questionnaire about the frequency of consumption of fish, meat, eggs, and beans (see Table 4.). Many participants responded to the question as follows. They ate fish 1-2 times a week, meat 3-4 times a week, eggs 3-4 times a week, and beans 1-2 times a week.

In addition, participants answered the following questions: 6.6% of participants ate meat every day, 23.0% of participants ate eggs every day, and 18.0% of participants ate beans every day. However, none of the participants ate fish every day.

Table 4. Results of a frequency survey of 61 participants eating fishs, meats eggs, and beans (%)

	1	2	3	4	5	6	7
	every day	5~6/week	3~4/week	1~2/week	2~3/month	1/month	rarely eat
Frequency of eating fishs	0.0	3.3	34.4	41.0	18.0	1.6	1.6
Frequency of eating meats	6.6	21.3	67.2	4.9	0.0	0.0	0.0
Frequency of eating eggs	23.0	18.0	47.5	11.5	0.0	0.0	0.0
Frequency of eating beans	18.0	13.1	29.5	34.4	4.9	0.0	0.0

Table 5 shows the results of a self-administered questionnaire survey of participants regarding the desired purchase price of commercially available fish that even the bones can be eaten. As a result, among the participants, the most requested price was 200 yen (about 1.5 US dollars). However, the reality is that the price of commercially available fish with edible bones in 400 yen (about 3-4 US dollars). It is about twice the purchase price requested by the participants.

Table 5. Desired purchase price of commercially available fish tha can eat up to bones
Average ± standard deviation = 266 ± 162 yen (Japanese yen)

Questions	50 yen	100 yen	120 yen	150 yen	180 yen	200 yen	250 yen	280 yen
Suggested purchase price (number)	1	5	1	8	1	16	5	2
Suggested purchase price (%)	1.6	8.2	1.6	13.1	1.6	26.2	8.2	3.3
Questions	298 yen	300 yen	350 yen	400 yen	500 yen	800 yen	1000 yen	no answer
Suggested purchase price (number)	1	7	2	5	4	1	1	1
Suggested purchase price (%)	1.6	11.5	3.3	8.2	6.6	1.6	1.6	1.6

IV. Discussions

From the results of a self-administered questionnaire survey conducted on 61 females, they did not feel a lack of protein, but felt a lack of calcium. However, 61 females wanted to consume both protein and calcium actively. The participants knew that even the bones of the fish were edible and tried to eat them, but the purchase price they wanted to buy them at a price far below the actual market price. Therefore, consumers consider fish that can be eaten, even the bones, expensive. As the protein in daily life, it is possible to purchase eggs, beans, and meat every day, but it is not easy to eat fish every day. Fish contain not only high-quality protein, but also large amounts of EPA and DHA1), which are suitable for blood vessels[2-4] and the brain[5]). In order to prevent Alzheimer's disease[6,7], it is desirable to take EPA and DHA, which are abundant in fish oil. They are also reported to be effective in preventing neuropathy[8] and leading to health promotion by numerous reports[9,10]. Eating fish for calcium[11] and for protein[12,13] intake is recommended. However, the high price of fish prevents consumers from purchasing fish daily. Even if it is challenging to buy raw fish, is it possible to increase the consumption of fish whose bones are edible by storing it at room temperature in retort pouches and using canned food?

Encourage consumers to consume processed fish caught in season (such as canned fish that can store for a long time and even the bones are edible) to increase protein and calcium intake.

V. Conclusions

Concerning the consumption of calcium, which did not meet the Japanese dietary intake standards, and the consumption of fish, which is declining a self-administered questionnaire survey was conducted on 61 females. As a result, the participants felt that their protein intake was sufficient, but felt that their calcium intake was low. The participants answered that they wanted to ingest both protein and calcium actively. The participants knew that there were fish available on the market that could be eaten, including the bones, from which calcium and protein could be ingested simultaneously, but they did not know that the price was higher than the asking price. Some participants ate eggs, meat, and beans daily, but none ate fish daily. In the future, we would like to introduce low-price canned fish and retort-pouch foods to consumers, increase the consumption of fish, even the bones which can be eaten, and increase the intake of calcium and protein.

Acknowledgements

This study was supported by the Japanese Society of Taste Technology, 2021.

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Global Journal of Medical Research: L
Nutrition & Food Science
Volume 22 Issue 2 Version 1.0 Year 2022
Type: Double Blind Peer Reviewed International Research Journal
Publisher: Global Journals Inc. (USA)
Online ISSN: 2249-4618 & Print ISSN: 0975-5888

Questionnaire Survey Results on Chewing of 90 People Who Participated in the Health Class

By Naomi Katayama
Nagoya Womens University

Abstract- Decreases in masticatory and swallowing ability are associated with age, but it is possible to maintain these functions through training. This research is the results of a questionnaire survey conducted on 90 participants (4 men and 86 women) in a health class held in 2021.

We conducted a questionnaire survey of 90 participants on chewing to understand the current situation. I cmpleted a 20-item questionnaire survey on chewing and swallowing. The results of the questionnaire are as follows.1) 4.4% with a removable partial denture, 2) 0.0% with complete dentures, 3) Currently attending a dentist 13.3%, 4) 36.7% undergoing regular dental examinations, 5) There are teeth currently being treated 6.7%, 6) The gums have been swollen 34.4%, 7) Blood comes out of the gums 38.9%, 8)34.4% who are usually interested in teeth, 9) Have heard the name of xylitol 97.8%, 10) Have listened to the name of mutants bacteria 36.7%, 11) Can bite apples with their skin 85.6%, 12) Confident in my teeth 20.0%, 13) My teeth are strong 41.1% 14) 8020 I am exercising 46.7%.

Keywords: questionnaire survey, chewing, time to eat.

GJMR-L Classification: DDC Code: 001.433 LCC Code: LB2823

Strictly as per the compliance and regulations of:

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Keywords: questionnaire survey, chewing, time to eat.

I. Introduction

In Japan, the 8020 campaign has been widely publicized by peoples. This campaign is that you should have 20 teeth and chew your teeth to eat enough to maintain your nutrition and health. If we finish eating in a shorter time than the signal reaches the satiety center, you may overeat. And our blood sugar level will rise sharply, and excess sugar will accumulate in our body as fat, resulting in obesity.

II. Materials and Methods

a) Participants

Participants were informed about the study, signed a consent form, and voluntarily participated in this study. A chewing questionnaire Survey was conducted on 90 people who participated in the health class. Participants voluntarily participated in the chewing questionnaire.

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b) Chewing questionnaire survey

Participants completed a self-administered questionnaire about twenty items related to chewing. Table 1 shows the contents of each item.

Table 1. Chewing questionnaire

20-item
Using removable partial dentures
Using full dentures
Currently attending a dentist
Have regular dental examinations
Have a tooth that is currently being treated
Have swollen gums
Blood comes out of the gums
Always interested in teeth
Heard the name of xylitol, which usually chews gum
Heard the name of Mutans bacteria, which usually chews gum
Can bite an apple with its skin
Have confidence in teeth
Think teeth are strong
Know the 8020 movement
There is a tooth that is an imsert tooth
Think I am chewing on food
Chew a bite of food more than 30 times
Do you brush your teeth after a snack?
Did you do fluoridation of your teeth when you were in elementary school?
habit of chewing gum

c) Questionnaire survey results regarding time spent on meals

Participants filled in a self-administered questionnaire for the time spent on the three meals.

d) Ethical review board

This study was conducted with the approval of the Ethical Review Board (Nagoya women's university 'hito wo mochii ta kennkyuu nikansuru iinnkai'). The approval number is 2020-26.

III. Results

a) Participant results

There were 90 participants, fourmales and 86 females. The age distribution is shown in Table 2.

Table 2. Age distribution of participants

	10s	20s	30s	40s	50s	60s
Maie	0	2	0	1	1	0
Female	25	42	4	7	6	2

Show the basic information of the participants in Table 3. The Average± standard deviation of age for 86 females is 26.8 ± 12.3. The Average± standard deviation of the age for four males is 35.8 ± 18.3.

Table 3. Basic information of participants (n=90)

	Female (n=86)			Male (n=4)		
	Age	Height	Weight	Age	Height	Weight
Average	26.8	157.0	50.7	35.8	172.0	66.3
Atandard diviation	12.3	5.4	6.8	18.3	5.7	5.3
Median	20	157.75	50	34.5	173.5	66.5
Maximum	69	169.6	80	54	177	72
Minimum	12	147	39	20	164	60

b) Questionnaire survey results

Table 4 shows the results of a questionnaire survey on mastication for 90 participants. Of the participants, 4.4% had partial dentures, and none had complete dentures. 36.7% of the participants went to the dentist regularly. Of the participants, 97.8% knew the name of Xylitol, and 46.7% knew the 8020 campaign (keeping 20 teeth at age 80). Among the participants, 47.8% felt they could chew food well, and 41.1% felt that their teeth were strong. Among the participants, 67.8% had their teeth treated with fluoride in elementary school, and 22.2% had the habit of chewing gum.

c) Questionnaire survey results regarding time spent on meals results

Most the participants spent 10 minutes on breakfast, 15 minutes on lunch, and 20 or 30 minutes on dinner.

The average time of participants was 12.8 minutes for breakfast, 18.6 minutes for lunch, and 24.8 minutes for dinner.

IV. Discussion

Most participants who attended the health class this time did not have a habit of chewing gum. However, almost everyone knew the word xylitol. But about half of the participants knew the talk of the 8020 campaign. Participants could bite the apple with the skin, but were less confident in the teeth. Participants replied that they chew food, but did not chew 30 times. If participants can prevented by eating the food bite little over time, the blood glucose level after a meal can moderate. Many researchers have reported the relationship between rumination and cognition/dementia risk[1)], and other effects on brain functions[2)]. Masticatory force is measured by device development and computational model[3,4)], and research to clarify occlusal ofrce[3,4,5,6)] reports. Studies have also reported that chewing gum increases bite force[7)]. The authors reported that high school students, university students, and middle-aged adults ate a meal less than 30 minutes per meal[8)]. In addition, the authors reported the results of training chewing power by chewing gum every day before meals for 30 days[9)]. Furthermore, the authors reported whether or not chewing gum improves masticatory strength[10)]. We want to convey the importance of chewing through these educational activities.

V. Conclusions

We reported the results of a self-administered questionnaire survey on mastication performed on 90 participants who voluntarily consented to the study. As a result, less than half of the participants answered that they consciously had solid teeth and could chew food well. The average time taken by the participants to eat every three meal was less than 30 minutes. Many of the participants had no habit of chewing gum.

Acknowledgements

This study was supported by the Japanese Society of Taste Technology, 2019.

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Global Journal of Medical Research: L
Nutrition & Food Science
Volume 22 Issue 2 Version 1.0 Year 2022
Type: Double Blind Peer Reviewed International Research Journal
Publisher: Global Journals Inc. (USA)
Online ISSN: 2249-4618 & Print ISSN: 0975-5888

Assessment of Body Mass Index (BMI) and General Health Status of Male Auto-Rickshaw Drivers in Garia, Kolkata

By Dr. Sampa Mitra

Abstract- *Background:* In India, auto-rickshaw is one of the main modes of public transport in urban and semi-urban areas; however, auto-rickshaw drivers often suffer from various nutritional deficiencies.

Objective: The objective of this work is to assess the body mass index (BMI) and the general health status of the auto-rickshaw drivers of the Garia Southern Avenue auto-rickshaw stand (in Kolkata).

Method: To achieve this objective, a cross-sectional study has been undertaken, whereby data have been collected in February-July 2018, regarding duration of work, nature of addiction, ownership pattern, general clinical status, BMI and body fat percentage, of 157 male auto-rickshaw drivers, attached to the aforesaid stand, and belonging to the age-group of 18-55 years. Subsequently, the binomial test has been conducted at 5% level of significance.

Keywords: *auto-rickshaw driver, Kolkata, body mass index, health status, binomial test, cross-sectional study.*

GJMR-L Classification: *DDC Code: 615.704 LCC Code: RM301*

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Results: The test results show that a significant number of auto-rickshaw drivers: (a) work for a long time (>8 hours/day but ≤12 hours/day), (b) are addicted to tobacco (both smoking and chewing forms), but not alcohol, (c) own the auto-rickshaw, (d) enjoy good health (except the occurrence of caries), and (e) have normal or higher than normal BMI and body fat percentage. Hence, this study shows that a significant number of auto-rickshaw drivers, attached to the aforesaid auto-rickshaw stand, have not-so-bad economic condition, and enjoy good health (except the occurrence of caries), despite long hours of work and addiction to tobacco; however, there is a possibility that, either they are overweight/obese currently, or they have a tendency towards being overweight/obese in the near future.

Keywords: auto-rickshaw driver, Kolkata, body mass index, health status, binomial test, cross-sectional study.

I. Introduction

In India, auto-rickshaw is one of the main modes of public transport in urban and semi-urban areas. In Kolkata also, auto-rickshaw services are there. However, auto-rickshaw drivers often suffer from various occupational hazards. Their lifestyle is not quite conducive to health, and they often experience irregularity of meals, among other things (1). Prolonged hours of work often leads to insufficient sleep and less physical activity (2). Besides, often there is a high prevalence of smoking and drinking among the auto-rickshaw drivers. All these factors may contribute to various health-related problems.

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The objective of this work is to assess the body mass index (BMI) and the general health status of the auto-rickshaw drivers of the Garia Southern Avenue auto-rickshaw stand (in Kolkata).

And, for this assessment, the binomial test has been conducted at 5% level of significance.

Before undertaking this study, a brief literature survey has been conducted. It has been found that a number of researchers have worked on the social condition, the economic status, and the health picture of the auto-rickshaw drivers in various cities of India. However, studies on the status of the auto-rickshaw drivers of Kolkata, are comparatively rare, and whatever articles I have come across, on this topic, do not cover the general health status of the auto-rickshaw drivers. As for example, one such work (by Agarwal et al.) focuses on the high prevalence of low back pain among the auto-rickshaw drivers of Kolkata (3). Now, the general health status of the auto-rickshaw drivers of Kolkata, is an important component of medical research, from which one may be able to draw significant and interesting inferences. And, this paper attempts to shed some light on this topic.

II. Materials and Methods

It is a cross-sectional study conducted between February 2018 and July 2018, on 157 male auto-rickshaw drivers, belonging to the age-group of 18-55 years; these drivers are attached to the Garia Southern Avenue auto-rickshaw stand. (The necessary research and ethical clearances have been taken from the institution to which I was attached during the study.) Only those auto-rickshaw drivers who are willing to participate, have been included in this study; and before inclusion, the nature and the purpose of the study have been explained to them in detail. In other words, informed consent has been taken from the auto-rickshaw drivers before including them in the study.

The sample size (s) has been calculated according to equation-1:

$$s = \frac{z^2 pq}{d^2} \qquad 1$$

where, z=value of the standard normal distribution corresponding to 95% confidence=1.96;

p=prevalence of malnutrition among adult Indian males=28.6%;
q=(100-p)=71.4%;
d=absolute error=10%.
After calculation, one will get s=78.41.
Considering a design effect of 2, the final sample size becomes 78.41 X 2=156.82≃157.

The sample (of size=157) has been collected on the basis of simple random sampling, from 1128 male auto-rickshaw drivers (belonging to the age-group of 18-55 years), associated with the Garia Southern Avenue auto-rickshaw stand.

A pre-designed and pre-tested questionnaire has been used to collect the relevant information from the auto-rickshaw drivers through interview (the questionnaire has been validated by pre-testing it among a few auto-rickshaw drivers belonging to the sample). Besides, the drivers have been subjected to thorough clinical examination.

Subsequently, the collected data have been tabulated, the binomial tests have been performed (at 5% level of significance) on them, and the results of the tests have been interpreted.

Binomial test is applied when an experiment has two possible outcomes viz., success and failure, and the probability of success is known. A binomial test is conducted to find out whether the observed result differs significantly from the expected one.

Here, the null hypothesis is that a significant number of auto-rickshaw drivers do not belong to the pertinent category, and the alternative hypothesis is that a significant number of auto-rickshaw drivers belong to the relevant category.

The formula (4) for obtaining the p-value (p1) is depicted in equation-2:

$$p1 = 2\frac{n!}{(n-X)!\,X!}p^{X}q^{(n-X)} \qquad 2$$

where, n=total number of auto-rickshaw drivers included in the study=157;
X=expected number of successes=n/2=78.5≃79;
p=observed proportion of success=proportion of auto-rickshaw drivers, belonging to a particular category;
q=observed proportion of failure=proportion of auto-rickshaw drivers, not belonging to the relevant category.

If p1<0.05, then it can be inferred that a significant number of auto-rickshaw drivers belong to the pertinent category (i.e., the null hypothesis is rejected and the alternative hypothesis is accepted); otherwise, the number of auto-rickshaw drivers, belonging to the relevant category, is not significant (i.e., the null hypothesis is accepted).

If p<0.5 (and consequently, q>0.5), then the value of p1 may result in wrong inference with regard to significance. Therefore, in such a situation (where p<0.5 and q>0.5), p and q are both taken as approximately (since n is an odd number) equal to 0.5, only for the sake of calculating p1.

The method employed in this work, is shown in fig.-1.

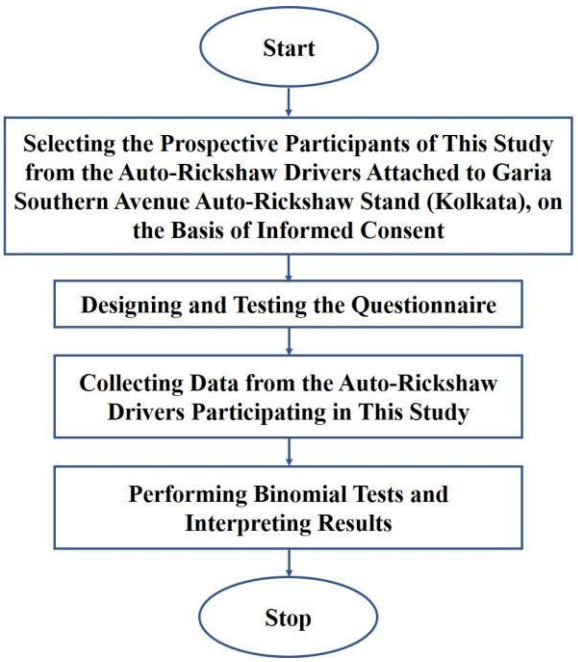

Fig.-1: Flowchart Depicting the Study Protocol

III. Results

The duration of work, nature of addiction, ownership pattern, general clinical status, and BMI and body fat percentage, of the auto-rickshaw drivers, are shown respectively in tables- 1, 2, 3, 4 and 5.

Table-1: Working status of the auto-rickshaw drivers (n=157)

Duration of work	Yes	No	p1
Working more than 6 days/week	87 (55.4%)	70 (44.6%) (number of drivers working 6 days/week)	0.0563
Working more than 8 hours/day (but not more than 12 hours/day)	104 (66.2%)	53 (33.8%) (number of drivers working 8 hours/day)	$2.9429X10^{-5}$

In table-1, the results of the binomial tests show that a significant number of auto-rickshaw drivers are working more than 8 hours/day (but not more than 12 hours/day), but the number of auto-rickshaw drivers, working more than 6 days/week (i.e., 7 days/week), is not significant.

Table-2: Addiction pattern of the auto-rickshaw drivers (n=157)

Addiction type	Yes	No	p1
Smoking	151 (96.2%)	6 (3.8%)	0
Chewing of tobacco	104 (66.2%)	53 (33.8%)	$2.9429X10^{-5}$
Consumption of alcohol	23 (14.6%) 79 (50.3%) (for calculating p1)	134 (85.4%) 78 (49.7%) (for calculating p1)	0.1274

In table-2, the outcomes of the binomial tests depict that a significant number of auto-rickshaw drivers have the habit of smoking and chewing tobacco, but the number of auto-rickshaw drivers who are addicted to alcohol, is not significant.

Table-3: Ownership status of the auto-rickshaw drivers (n=157)

Ownership	Yes	No	p1
Owner of the auto-rickshaw	100 (63.7%)	57 (36.3%)	$3.6747X10^{-4}$

In table-3, the binomial test result shows that a significant number of auto-rickshaw drivers own the auto-rickshaw.

Table-4: General clinical assessment of the auto-rickshaw drivers (n=157)

Criterion	Yes	No	p1
Good appearance	157 (100%)	0 (0%)	0
Normal angles of mouth (i.e., no ulcer at the angles of the mouth)	157 (100%)	0 (0%)	0
Normal tongue colour	136 (86.6%)	21 (13.4%)	$2.2058X10^{-27}$
Normal gum	146 (93.0%)	11 (7.0%)	$6.2128X10^{-47}$
Absence of fluorosis in teeth	157 (100%)	0 (0%)	0
Teeth with caries	99 (63.1%)	58 (36.9%)	$6.2498X10^{-4}$
Normal hair condition	157 (100%)	0 (0%)	0
Normal skin appearance	143 (91.1%)	14 (8.9%)	$1.6591X10^{-39}$
Absence of oedema	157 (100%)	0 (0%)	0
Absence of pallor	136 (86.6%)	21 (13.4%)	$2.2058X10^{-27}$

In table-4, according to the outcomes of the binomial tests, a significant number of auto-rickshaw drivers have good appearance, normal angles of mouth, normal tongue colour, normal gum, no fluorosis in teeth, normal hair condition, normal skin appearance, no oedema, and no pallor. However, the binomial test results, in table-4, also show that the number of auto-rickshaw drivers, having teeth with caries, is significant.

Table-5: BMI and body fat percentage of the auto-rickshaw drivers (n=157)

Status	Yes	No	p1
BMI (in kg/m^2) in or above normal range (18.50-24.99)	127 (80.9%)	30 (19.1%)	$1.0328X10^{-17}$
Normal or higher than normal body fat percentage (body fat percentage data have been collected using body fat analyser)	156 (99.4%)	1 (0.6%)	0

In table-5, as per the results of the binomial tests, a significant number of auto-rickshaw drivers have BMI in or above the normal range (18.50 kg/m^2 – 24.99 kg/m^2), and normal or higher than normal body fat percentage.

IV. Discussions

The following inferences can be drawn from tables- 1, 2, 3, 4 and 5:

- A significant number (66.2%) of auto-rickshaw drivers, attached to the Garia Southern Avenue auto-rickshaw stand, work for a long time.
- A significant number of drivers are addicted to tobacco (both smoking (96.2%) and chewing (66.2%) forms), but not alcohol.
- A significant number (63.7%) of drivers own the auto-rickshaw; this fact indicates that the economic condition of a significant number of drivers is not bad (it is a qualitative idea).
- A significant number of drivers enjoy good health (except the occurrence of caries) (as per the general clinical assessment), in spite of having long working hours. This is most probably because of their not-so-bad economic condition which allows them to get sufficient food of acceptable quality.
- A significant number of drivers have normal or higher than normal BMI (80.9%) and body fat percentage (99.4%). Thus, there is a possibility that a significant number of drivers is either overweight/obese currently, or likely to become overweight/obese in the near future. This is most probably because of their not-so-bad economic condition (as stated above), and also the nature of their occupation (which demands the drivers to remain in sitting position for a long time).

Hence, this study shows that a significant number of auto-rickshaw drivers, attached to the Garia Southern Avenue auto-rickshaw stand, have not-very-bad economic condition, and enjoy good health (except the occurrence of caries), despite long hours of work, and addiction to tobacco; and, a significant majority of them are either overweight/obese currently, or likely to become overweight/obese in the near future.

Now, it will be prudent to take a look at the findings of some of the other researchers working on auto-rickshaw drivers.

Yesurajan et al. found that smoking, alcohol abuse, and obesity are some of the common health risk factors of the auto-rickshaw drivers of Madurai (5); in the current study, however, alcohol abuse is not a problem for a significant number of drivers (only 14.6% have been found to consume alcohol).

Gupta et al. undertook a study on the auto-rickshaw drivers of Mumbai, and found that only 15% of the study population are smokers (6); in contrast, I have found that a significant number (96.2%) of auto-rickshaw drivers are addicted to smoking.

Chougule et al. worked on the auto-rickshaw drivers of Kolhapur, and found that most of them enjoy good health (7); this finding is similar to what I have observed in the current work.

The study of Debbarma et al. on the auto-rickshaw drivers of Agartala, shows that majority of the study population (73.81%) were suffering from caries (8); I have also found that a significant number (63.1%) of auto-rickshaw drivers have teeth with caries.

Jain et al. conducted research on the auto-rickshaw drivers of Gwalior, and found that among the study population, prevalence of overweight was 26 % and central obesity was 6% (9); on the contrary, in the current study, the results indicate that a significant number of drivers are either overweight/obese currently, or likely to become overweight/obese in the near future.

V. Conclusions

This study has attempted to assess the BMI and the general health status of the auto-rickshaw drivers, attached to the Garia Southern Avenue auto-rickshaw stand, by collecting data regarding duration of work, nature of addiction, ownership pattern, general clinical status, BMI and body fat percentage, of the auto-rickshaw drivers. From the results of the binomial

tests conducted on these data, it can be concluded that a significant number of auto-rickshaw drivers enjoy not-so-bad economic condition, and good health, in spite of having long working hours, and being addicted to tobacco; however, there is a probability that they are either overweight/obese currently, or likely to become overweight/obese in the near future.

A plus point of this study is that it has been able to draw the above conclusions without performing any expensive and/or complicated medical examination.

However, if data regarding the respiratory system, the musculo-skeletal system, and some general health parameters like blood pressure and blood sugar level, of the auto-rickshaw drivers, were also collected and analysed, a more or less comprehensive idea regarding the health status of the drivers could have been obtained. Also, if auto-rickshaw drivers from other auto-rickshaw stands were also included in the study, it would have yielded a more general picture regarding the health status (of the auto-rickshaw drivers of Kolkata). If possible, these assignments can be taken up in future.

Acknowledgements

The author is deeply indebted to Ms. Poulami Basu, MSc (Applied Nutrition) student (session: 2016-18), AIIH & PH, Kolkata, for collecting the data and helping in the analysis.

Besides, the author wants to express her gratitude to the auto-rickshaw drivers (who have participated in this study), attached to the Garia Southern Avenue auto-rickshaw stand, for their cooperation in the data collection process.

Source of Funding: Personal Source

Conflict of Interest: The author certifies that there is no conflict of interest, involved in this study.

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Global Journal of Medical Research: L
Nutrition & Food Science
Volume 22 Issue 2 Version 1.0 Year 2022
Type: Double Blind Peer Reviewed International Research Journal
Publisher: Global Journals Inc. (USA)
Online ISSN: 2249-4618 & Print ISSN: 0975-5888

The Effects of Early-Harvest Extra Virgin Olive Oil on Cognition and Mental Health of Primary (PPMS) or Secondary (SPMS) Progressive Multiple Sclerosis Patients

By Thanos Chatzikostopoulos, Magdalini Tsolaki, Greta Wozniak, Emmanouela Basgiouraki, Iordanis Saoulidis, Dimitrios Michmizos & Efrosyni Koutsouraki

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Abstract- *Aim of the study:* Over the last years the cognitive and mental health impairment in Multiple Sclerosis (MS) are indicated as important clinical symptoms in the course of the disease. Every beneficial therapeutic management with this target could lessen the disability caused by the disease and improve the quality of life of MS patients. It is known that Extra Virgin Olive Oil (EVOO) can exert positive effects on cognition regarding neurodegenerative diseases. Phenolic compounds in EVOO have antioxidative and anti-inflammatory effects on the brain but all the mechanisms are not clear yet. The present pilot study examines the benefits of early harvest EVOO (EH EVOO) on cognition and mental health regarding MS.

Keywords: *early harvest extra virgin olive oil, cognition, mental health, multiple sclerosis.*

GJMR-L Classification: *DDC Code: 616.8914 LCC Code: RC455.4.L67*

Strictly as per the compliance and regulations of:

The Effects of Early-Harvest Extra Virgin Olive Oil on Cognition and Mental Health of Primary (PPMS) or Secondary (SPMS) Progressive Multiple Sclerosis Patients

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Abstract- Aim of the study: Over the last years the cognitive and mental health impairment in Multiple Sclerosis (MS) are indicated as important clinical symptoms in the course of the disease. Every beneficial therapeutic management with this target could lessen the disability caused by the disease and improve the quality of life of MS patients. It is known that Extra Virgin Olive Oil (EVOO) can exert positive effects on cognition regarding neurodegenerative diseases. Phenolic compounds in EVOO have antioxidative and anti-inflammatory effects on the brain but all the mechanisms are not clear yet. The present pilot study examines the benefits of early harvest EVOO (EH EVOO) on cognition and mental health regarding MS.

Materials and Methods: The participants had been diagnosed with primary (PPMS) or secondary (SPMS) progressive MS and they were evaluated using a neuropsychological assessment, which covers a wide range of cognitive and mental health functions before and after one year of treatment.

Results: After one year of EH EVOO consumption the results indicated that the patients showed significant improvement in processing speed (p=.01), visuospatial memory (p=.002) and functions related to the frontal lobes, such as mental flexibility and adaptation to the environment (p=.017). On the contrary, patients, who were not consuming EH EVOO (control group), did not show significant improvement neither in processing speed (p=.443) or functions related to the frontal lobes (p=.357).

Conclusions and Clinical Implications: The consumption of EVOO can be helpful for some cognitive functions, such as visuospatial memory and processing speed. For this reason, EVOO may have an important role in neuroprotection and neurodegeneration in MS patients.

Keywords: early harvest extra virgin olive oil, cognition, mental health, multiple sclerosis.

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I. Introduction

According to National Multiple Sclerosis Society Multiple Sclerosis (MS) can be defined as an immune-mediated process in which an abnormal response of the body's defense system is directed against the central nervous system (CNS). In this way, the immune system can precipitate neuroinflammation that, in turn, leads to demyelination and, subsequently, to axonopathy and neurodegeneration. Because of these damages to the CNS, numerous neurological symptoms may be occurred with severity that differs among MS patients [1].

The diagnosis of MS is based on international diagnostic criteria, although there is a great probability of false diagnosis due to many neurodegenerative diseases mimicking MS symptoms. According to the Revised McDonald Criteria (2017) the use of brain Magnetic Resonance Imaging (MRI) and cerebrospinal fluid (CSF) analysis can expedite this process by confirming the damages. Besides these tests, the presence of oligoclonal bands in the CSF can confirm the diagnosis.

The International Advisory Committee on Clinical Trials of MS in 2013 has defined four types of MS: clinically isolated syndrome (CIS), relapsing-remitting MS (RRMS), secondary progressive MS (SPMS) and primary progressive MS (PPMS). Specifically, SPMS consists of an initial relapsing-remitting course, which will be evolved to a progressive disability. Furthermore, SPMS can be defined as active, if there is evidence of new MRI activity, or no active, as well as worsening, if there is a confirmed increase of disability after a relapse or not worsening. PPMS does not include relapses and remissions, but the neurological functions get worse gradually after the first symptoms. The same classification is applied in this type too [2]. For every clinical attack approximately 10 "asymptomatic" lesions are noted on MRI [3].

In 5%-15% of cases there is a primary progressive onset (PPMS) typically with gradual increase of disability on one dominant neuronal system.

Some of the commonest symptoms are progressive spastic paraparesis, sensory ataxia, cerebral ataxia, cognitive and visual progressive decline [3]. Remyelination can be seen in all stages of the disease but most commonly in its progressive types. [2]

a) Cognitive and mental health impairment in MS

Over the last two decades, cognitive impairment has been recognized as an important factor that affects MS patients' quality of life [4]. It is proved that it affects up to 65% of patients, and yet cognitive testing remains uncommon, due to the lack of simple and reliable tests that have been validated for the MS population. For instance, the Mini Mental State Examination (MMSE) which is used to detect cognitive deficits due to dementia, is deemed not to be appropriate for MS population [4,5]. Cognitive impairment can precipitate in all forms and stages of MS presenting with a variation of symptoms and a range of severity that differs among the patients [5].

At first, memory and especially free recall of verbal and visuospatial material seems to be affected [6]. Then, working memory and attention reportedly get impaired, because MS patients demonstrate reduced performance in complex attention tasks [7]. Executive functions, such as planning, problem-solving and self-monitoring are, also, declined [8]. Nevertheless, one of the most common features of cognitive impairment in MS patients is reduced processing speed, manifested as longer reaction time to stimuli and reduced speed of memory scanning [9,10].

Some risk factors have been identified, such as age, the subtype of the disease, the disease's duration, the characteristics of the pathological lesions, progressive fatigue, the prescribed medication, as well as depression and anxiety disorders. Moreover, the fact that the patients may be aware of their cognitive decline can compound their negative attitude, which can lead the patients overestimating their cognitive deficits and being overwhelmed by them. For this reason, early detection is crucial to prevent further decline [5].

b) The benefits of EVOO

There is evidence that EVOO has neuroprotective effects against aging and neurodegenerative diseases, such as prodromal stages of Alzheimer's disease. Phenolic compounds in olive oil have been found to exert positive effects on inflammatory markers, as well as cellular and neuropsychological functions [11]. Furthermore, Ruano et al (2005) presented that consumption of EVOO can lower markers of oxidative stress such as F2-isoprostane [12].

The mechanisms behind EVOO's neuroprotective effects are not yet crystallized. It is primarily known that it has antioxidative effects, because it contains antioxidant molecules and free radical scavengers, which neutralize the toxic moieties and scavenge many endogenous and exogenous free radicals and oxidants. Moreover, EVOO has anti-inflammatory effects because its compounds inhibit many inflammatory mediators. Furthermore, EVOO is known regarding its anti-apoptotic properties. In other words, besides reducing toxins, oxidative stress and hypoxia, olive oil consumption, also, inhibits programmed cell death, called apoptosis, which is a very important parameter in neurodegenerative diseases [11]. Many parameters contribute to the effects of these compounds, such as their concentration and the extent of their absorption and metabolism [13].

c) Objectives

Taking into consideration that there are only a few published studies on the benefits of EVOO in cognition and mental health, the primary objective of this pilot study is to view the effects of EVOO in cognition and mental health of patients with progressive types of MS by using extensive neuropsychological assessment and evaluating participants' cognition and mental health for one year. Our hypothesis, therefore, was that the MS patients would show progressive improvement in their post-therapy assessments.

II. Materials and Methods

a) Participants

The participants had the diagnosis of PPMS and SPMS and they were not receiving specific MS medication because previous treatments had failed. Twenty patients (12 women and 8 men aged 35-65 years old) took part in the present study as an intervention group and ten patients (7 women and 3 men) as a control group (Table 1). The patients were assigned randomly in each group and there were not statistically significant differences between them regarding gender, education and age.

Table 1: Demographics in both group of patients with MS

	Gender		Education		Age	
	Men	Women	M	Std	M	Std
Experimental group	12	8	15.25	9.031	47.85	11.726
Control group	7	3	12.80	4.237	41.50	14.470
p	0.592		0.436		0.367	

b) Procedure

All study participants read the information sheet and signed an informed consent stating that the research group have the permission to use their demographic data, which would be anonymized, such as gender, age and education, as well as their performance in the neuropsychological tests, for research purposes. Before the administration of EH EVOO, the participants were evaluated using an extended neuropsychological assessment which includes measurements that cover a wide range of cognitive functions and are mentioned below. The neuropsychological assessment took place in a soundproof room in 1st Department of Neurology of Aristotle University of Thessaloniki in Greece by a trained psychologist. Patients were instructed to take three tablespoons of EH EVOO per day. After six months and after one year they were called in for re-evaluation, using the same measurements but with alternative forms, wherever it was possible. The protocol of the present study is registered ClinicalTrials.gov with ID NCT04120675.

The EH EVOO, which was distributed to the participants, is Eliama D. V. Gold Health Claim High Phenolic Extra Virgin Olive Oil and contains high concentrations of polyphenol compounds (Oleocanthal, Oleacein, Oleuropein, Tyrosol & Hydroxytyrosol derivatives > 1200 mg /kg EVOO and very high concentration of Squalene 6800 mg and Tocoferol a). Eliama Daily Value Gold brings the certification of Health Claim. The health claim: The claim may be used only for olive oil which contains at least 5 mg of hydroxytyrosol and its derivatives (e.g. oleuropein complex and tyrosol) per 20g of EVOO. In order to bear the claim information shall be given to the consumer that the beneficial effect is obtained with a daily intake of 20g of EVOO et least. "According to European Regulation 432/2012 (L136/25.5.2012 p. 26), the Health Claim is brought only by the olive oil that contains at least 250gr. polyphenols per 1Kg. of olive oil. A chemical analysis of the product was done by an approved chemical lab, but also the polyphenols analysis from the Pharmacology Department of National Kapodistrian University of Athens.

c) Neuropsychological examination

1. *The Frontal Assessment Battery (FAB)* consists of 6 tasks and evaluates frontal deficits. These tasks can disclose deficits in conceptualization (similarity test), mental flexibility (lexical fluency), motor programming (Luria's fist-edge-palm, conflicting instructions, Go/ No go tests) and environmental autonomy (prehension behavior). So, it was useful in order to evaluate cognitive deficits which may have occurred due to frontotemporal lesions. The mean score for MS patients is 16.8 (SD=2) [14].
2. *The Brief International Cognitive Assessment for Multiple Sclerosis (BICAMS)* is a neuropsychological battery which has been validated in the Greek MS population and consists of the Symbol Digit Modalities Test (SDMT), the California Verbal Learning Test (CVLT) and the Brief Visuospatial Memory Test-Revised (BVMT-R) [15]. SDMT is used to detect deficits in visual processing speed. It consists of a series of nine symbols, each in association with a single digit. Participants enunciate the digit related to each symbol in a randomized sequence, as fast as they can, in a 90 seconds session. The total score is derived from the number of correct answers enunciating the digit related to each symbol in a randomized sequence, as fast as they can, in 90 s. Instead of the CVLT, the Greek adaption of this test (GVLT) was used in order to measure auditory and verbal memory. Participants have to learn as many words as they can from a list with 16 words and this procedure is repeated five times. Form A was used for the assessment and form B for the retesting session. The total score equals to the total number of recalled words [16]. BVMT-R measures visual and spatial memory. In this test, participants must remember a page of six shapes which is presented for ten seconds and this procedure is repeated three times. Each shape is scored with zero, one or two points, depending on accuracy and location. The sum of the scores of the three trials is the total score of the test. [17].
3. *The Perceived Deficits Questionnaire* is a self-reporting questionnaire measuring subjective cognitive deficits and is designed specifically for MS patients. It covers the cognitive functions which are most often impaired in MS and it concludes four subscales: attention, retrospective memory, prospective memory and planning/organizing. The total score is computed by the sum of raw scores for all items and it can range from 0-80, with higher scores indicating greater perceived cognitive impairment [18].
4. *The Mental Health Inventory (MHI)* is a reliable measure for patients' emotional condition. It consists of 18 items which are separated in four scales: anxiety, depression, behavioral control and positive affect. The total scores range from 0-100, with higher scores indicating better mental health [19].
5. *The Beck Depression Inventory (BDI)* is used to measure depression. It is designed to examine both somatic and cognitive aspects of depression and the Greek version has been validated previously [20] and has been widely used to date. The BDI is a 21-item self-reporting scale scored on a 4-point

scale (0-3). It has been shown to have good psychometric properties with test-retest correlations >0.90 in different studies. Moreover, it has shown satisfactory validity with agreement between BDI and psychiatrists' ratings of 56% and has also been shown sensitive to distinguish between depression and anxiety. Moreover, factor analysis generally reveals three inter-correlated factors indicating severity of depression. Scoring of the questionnaire is as follows: 0–9 no depression, 10–15 mild, 16–23 moderate, and 24–63severe depression [21]. The BDI has been validates for MS patients too and it is proved to be applicable for the evaluation of depression in this population [22, 23]

d) Statistics

Statistical analysis was performed with the use of SPSS 25 Statistical package. A Wilcoxon Signed-Ranks Test was conducted in order to calculate the score differences in neuropsychological assessments before and after 1 year of therapy (intragroup comparisons). Moreover, an Independent Samples T-test was conducted to compare the means of the two groups before and after EH- EVOO's consumption (intergroup comparisons).

III. Results

At first, the differences between the two groups were calculated before the beginning of therapy, using Independent Samples T-test (Table 2). For this reason, the results of the first neuropsychological assessment of the two groups were compared in order to be insured that the two groups start from the same baseline. The results indicated that there were not statistically significant differences between the two groups and the Cohen's d test confirmed that the sample's size does not affect the results.

Table 2: Means' comparison between the two groups before the beginning of therapy using Independent Samples T-test

	Intervention group (n=20)		Control group (n=10)				
	M	Sd	M	Sd	t(30)	p	Cohen's d
FAB	16.00	1.806	15.00	2.494	-1.258	.219	.000459
GVLT	48.85	9.167	52.60	15.467	.837	.410	.4767
BVMT	22.85	5.373	22.00	5.249	-.411	.684	.4261
SDMT	31.70	7.540	30.00	11.785	-.481	.634	.0316
PDQ	5.15	3.746	4.90	2.601	-.189	.852	.0136
BDI	9.45	5.336	6.20	5.287	-1.577	.126	.0774
MHI	69.45	9.950	69.80	19.657	.065	.948	.0022

*Significance levels: *p< .05*
***p< .01*

An Independent Sample T-test was, also, conducted to compare the means of the two groups after one year of EVOO's consumption (Table 3) and there were some statistically significant results (Table 3). At first, in the FAB there was statistically significant difference between experimental group (M=17.55, Sd=0.759) and control group (M=15.30, Sd=3.164), t (30)=-3.058, p<.01. Secondly, there were differences in the two subtests of BICAMS, the BVMT and the SDMT. In the BVMT there was statistically significant difference between experimental group (M=26.35, Sd=4.056) and control group (M=20.50, Sd=5.442), t(30)=-3.321, p<.01. In the SDMT there was, also, statistically significant difference between experimental group (M=36.80, Sd=6.023) and control group (M=29.80, Sd=7.642), t(30)=-2.744, p<.05.

Table 3: Means' comparison between the two groups after one year therapy using Independent Samples T-test

	Intervention group		Control group				
	M	Sd	M	Sd	t(30)	p	Cohen's d
FAB	17.55	.759	15.30	3.164	-3.058	.005**	.0978
GVLT	52.90	7.567	51.10	14.888	-.443	.661	.0152
BVMT	26.35	4.056	20.50	5.442	-3.321	.002**	.1212
SDMT	36.80	6.023	29.80	7.642	-2.744	.010*	.1017
PDQ	4.25	3.007	6.30	2.497	1.856	.074	.0742
BDI	6.35	3.281	8.60	8.017	1.099	.281	.0367
MHI	77.35	4.987	67.70	20.618	-2.011	.054	.0643

*Significance level: *p< .05*
***p< .01*

As far as the intragroup comparisons (Table 4), the functions that are related to the frontal lobes were improved. Specifically, the FAB scores compared before and after therapy: Fourteen (14) out of twenty patients performed better after therapy. A Wilcoxon Signed-Ranked Test indicated that this difference was statistically significant, z=-3.329 p<.01. BICAMS scores, also, indicated improvement in specific cognitive functions. There were not statistically significant differences in GVLT score but there was statistically significant improvement in BVMT and SDMT scores. In the BVMT fifteen (15) out of twenty patients performed better after therapy and a Wilcoxon Signed-Ranked Test statistically significant improvement, z=-3.170, p<.01. In the SDMT, scores before and after therapy indicated statistically significant differences because seventeen (17) patients performed better after therapy than before, z=-3.467 p<.01.

As far as depression and negative emotions, great improvement was indicated by both the BDI and the MHI. Specifically, according to BDI, depressive symptoms were improved in 16 patients after therapy compared to their previous scores, z=-3.523 p< .01 Furthermore, MHI indicated statistically significant improvement in patients' general mental health, because 17 patients had less mental health problems after therapy than before, z=-3.456p< .01.

Table 4: Calculation of differences before and after one one year therapy with EH-EVOO using Wilcoxon Signed-Ranks Test

Before/ after	Negative ranks			Positive Ranks			Test Statistics		
	n	Mean rank	Sum of ranks	n	Mean rank	Sum of ranks	Ties	Z	p
FAB	0	.00	.00	14	7.50	105.00	6	-3.329	.0001**
GVLT	9	5.39	48.50	9	13.61	122.50	2	-1.615	.106
BVMT	3	4.33	13.00	15	10.53	158.00	2	-3.170	.002**
SDMT	1	6.00	6.00	17	9.71	165.00	2	-3.467	.0001**
PDQ	5	5.10	25.50	8	8.19	65.50	7	-1.419	.156
BDI	2	2.50	5.00	16	10.38	166.00	2	-3.523	.0001**
MHI	3	4.17	12.50	17	11.62	197.50	0	-3.456	.001**

*Significance levels: *p<.05*
***p<.01*

However, in control group there was no statistically significant changes (Table 5). These results of control group in combination with the statistically significant results of intervention group confirm EVOO's benefits in certain sectors of cognition.

Table 5: Calculation of differences before and after one year without therapy using Wilcoxon Signed-Ranks Test

Before/ after	Negative ranks			Positive ranks			Test Statistics		
	n	Mean rank	Sum of ranks	n	Mean rank	Sum of ranks	Ties	Z	p
FAB	3	4.83	14.50	5	4.30	21.50	2	- .496	.620
GVLT	6	4.50	27.00	3	6.00	18.00	1	-.539	.590
BVMT	6	4.83	29.00	2	3.50	7.00	2	-1.611	.107
SDMT	3	6.33	19.00	5	3.40	17.00	2	-.140	.888
PDQ	7	5.71	40.00	2	2.50	5.00	1	-2.111	.035*
BDI	6	4.50	27.00	1	1.00	1.00	3	-2.111	.027*
MHI	5	4.90	24.50	4	5.13	20.50	1	-.237	.813

*Significance levels: *p<.05*
***p<.01*

IV. Discussion

The aim of the present study was to detect and quantify the benefits of EH EVOO in protecting the cognition and mental health of MS patients. The hypothesis was that, because of the protective effects already attributed to EH EVOO consumption, the patients from the intervention group would demonstrate improved scores in their neuropsychological assessment after six months of EH-EVOO treatment. In general, the results showed that in the intervention group there was statistically significant improvement in the FAB, the BVMT and the SDMT. These results show that EH EVOO has positive effects in executive functions, visual memory and processing speed. At the same time, there were no statistically significant results regarding the control group. Although there are no studies about the benefits of EH EVOO in MS, there are studies supporting that EVOO (and the Mediterranean diet) can prevent cognitive decline and Alzheimer's disease in the elderly population [24, 25]. So, EVOO may have an important role in neuroprotection and staving off neurodegeneration, even if there is still a need for more studies regarding MS and other neurodegenerative diseases, such as Parkinson's disease, ALS etc.

With the use of FAB, functions related to the frontal lobes were evaluated. The fact that the patients had statistically significant improvement in FAB means that the EH EVOO may be helpful in order to improve goal directed behaviors, mental flexibility and adaptation to the environment. Moreover, the statistically significant scores in SDMT may be indicative of improvement in the brain processing speed. These findings confirm that some cognitive functions, which are impaired due to the neurodegeneration of MS, can be improved with consumption of EH EVOO. However, the benefits are not limited to MS patients.

A three-city study has already claimed the beneficial effects of EVOO on cognition. This study was conducted in three French cities and used neuropsychological assessments repeated every two years to measure any cognitive decline and assess risk factors for dementia's symptoms. The results indicated that participants who were less luckily of demonstrating cognitive deficit for verbal fluency and visual memory, whereas, during the 4-year follow-up there was significant association between intensive use of olive oil and prevention of visual memory's decline [26].

In the present study, verbal fluency was evaluated with the second task of FAB and visual memory was evaluated by the BVMT. So, regarding cognitive decline, our extensive study expands the above findings, adding that EH EVOO is beneficial in these fields for MS patients too.

EVOO offers protection to neuronal functions in neurodegenerative diseases as well. Olive oil's phenolic compounds contain natural antioxidants, including vitamins E, which may reduce neuronal damage and death from oxidative reactions by inhibiting the generation of reactive oxygen species, apoptosis, protein oxidation, damages to cell membranes and β-amyloid toxicity. However, the mechanisms, which are used in order to achieve these benefits, are not clear yet and behest further study [11].

Another extensive study, the PREDIMED-NAVARRA randomized trial, which examined the benefits of Mediterranean diet, supplemented with EVOO, on people with high vascular risk, also advocate the present study's results. In this study, the neuropsychological assessment included Mini Mental State Examination (MMSE) and Clock Drawing Test (CDT), which evaluate cognitive deficits and cover a wide range of cognitive functions. The results of these assessments indicated that after 6,5 years of follow-up the participants had better global cognitive performance and supported the protective effects of Mediterranean diet with EVOO on cognitive function [27].

As far as patients' mental health, significant improvement was found in both BDI and MHI. Specifically, the majority of patients (16 out of 20) had lower scores in BDI, which means that the patients had fewer depressive symptoms after six months of using EH EVOO. In MHI, the majority of patients (17 out of 20) had higher scores after consumption of EVOO, which means that they had less mental health problems and this was confirmed by the findings of the BDI. Observational studies confirm these results because they have pointed to an inverse association between adherence to Mediterranean diet (MeDi) and risk for depression. Furthermore, two clinical trials have demonstrated significant improvement regarding depressive symptoms in patients who were following MeDi [17]. The PREDI-DEP trial was the first randomized clinical trial, designed to examine the role of the MeDi supplemented with EVOO in the prevention of recurrent depression. This study confirmed the positive effects of MeDi, in general, and EVOO in particular, in depression as it was found that they can reduce the recurrence of depression and increase the patients' quality of life [28]. Moreover, other studies have pointed that a low-fat diet supplemented with EVOO can reduce the physical and emotional disease burden in MS patients [29, 30]. A possible mechanism behind the benefits of EVOO in mental health is that it can lower the markers of the above-mentioned oxidative stress, such as F2-isoprostane [11, 12,].

A limitation of this pilot study is that apart from the three subtests of the BICAMS, and the BDI, the neuropsychological tests, which have been used, are not validated for the Greek MS population. However, this

research is ongoing, and it will be continued for years to come. So, these specific neuropsychological tests will be validated for the Greek population soon. Another limitation is the limited number of participants (n=30) because this is a pilot study testing the effects of EH EVOO, the feasibility of the present research protocol.

There had been no previous evidence presented about the benefits of EH EVOO, and olive oil in general, in protecting the cognition and mental health of patients with MS. So, the innovation of the present pilot study is that these results can expand the research in this field and encourage the use of EVOO in holistic treatments of MS. For this reason, studies with increased sample size and even more bold approaches will be useful confirming the present study's results and identifying the specific mechanisms by which olive oil offers its benefits.

Declarations

Acknowledgements: We would like to offer our special thanks to Family Company Ellis farm (https.//ellis-farm.com/) for the Donation of the Health Claim EVOO High Phenolic Early Harvest Eliama D.V. Gold as on this important research project. Furthermore, many thanks for Dr Prokopis Magiatis. Associate Professor at Faculty of Pharmacy, National and Kapodistrian University of Athens (Laboratory of Pharmacognosy and Natural Products Chemistry) for all certificates of EVOO.

Funding: The present study and the authors were not funded.

Conflicts of interest: The authors declare that they have no conflict of interest.

Ethics approval: The study protocol has been approved by Bioethics Committee of Greek Association of Alzheimer's Disease and Related Disorders.

Consent to participate: All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the Declaration of Helsinki 1964 and was approved by the local ethics committee. All the participants gave informed consent prior to their inclusion in the study.

Consent for publication: All the authors have consented for the publication of the study.

Availability of data and material: Data available upon duly justified request.

Conflicts of interest: none

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4. Manuscript to be submitted must include keywords, an abstract, a paper title, co-author(s') names and details (email address, name, phone number, and institution), figures and illustrations in vector format including appropriate captions, tables, including titles and footnotes, a conclusion, results, acknowledgments and references.
5. Authors should submit paper in a ZIP archive if any supplementary files are required along with the paper.
6. Proper permissions must be acquired for the use of any copyrighted material.
7. Manuscript submitted *must not have been submitted or published elsewhere* and all authors must be aware of the submission.

Declaration of Conflicts of Interest

It is required for authors to declare all financial, institutional, and personal relationships with other individuals and organizations that could influence (bias) their research.

Policy on Plagiarism

Plagiarism is not acceptable in Global Journals submissions at all.

Plagiarized content will not be considered for publication. We reserve the right to inform authors' institutions about plagiarism detected either before or after publication. If plagiarism is identified, we will follow COPE guidelines:

Authors are solely responsible for all the plagiarism that is found. The author must not fabricate, falsify or plagiarize existing research data. The following, if copied, will be considered plagiarism:

- Words (language)
- Ideas
- Findings
- Writings
- Diagrams
- Graphs
- Illustrations
- Lectures

- Printed material
- Graphic representations
- Computer programs
- Electronic material
- Any other original work

Authorship Policies

Global Journals follows the definition of authorship set up by the Open Association of Research Society, USA. According to its guidelines, authorship criteria must be based on:

1. Substantial contributions to the conception and acquisition of data, analysis, and interpretation of findings.
2. Drafting the paper and revising it critically regarding important academic content.
3. Final approval of the version of the paper to be published.

Changes in Authorship

The corresponding author should mention the name and complete details of all co-authors during submission and in manuscript. We support addition, rearrangement, manipulation, and deletions in authors list till the early view publication of the journal. We expect that corresponding author will notify all co-authors of submission. We follow COPE guidelines for changes in authorship.

Copyright

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Appealing Decisions

Unless specified in the notification, the Editorial Board's decision on publication of the paper is final and cannot be appealed before making the major change in the manuscript.

Acknowledgments

Contributors to the research other than authors credited should be mentioned in Acknowledgments. The source of funding for the research can be included. Suppliers of resources may be mentioned along with their addresses.

Declaration of funding sources

Global Journals is in partnership with various universities, laboratories, and other institutions worldwide in the research domain. Authors are requested to disclose their source of funding during every stage of their research, such as making analysis, performing laboratory operations, computing data, and using institutional resources, from writing an article to its submission. This will also help authors to get reimbursements by requesting an open access publication letter from Global Journals and submitting to the respective funding source.

Preparing your Manuscript

Authors can submit papers and articles in an acceptable file format: MS Word (doc, docx), LaTeX (.tex, .zip or .rar including all of your files), Adobe PDF (.pdf), rich text format (.rtf), simple text document (.txt), Open Document Text (.odt), and Apple Pages (.pages). Our professional layout editors will format the entire paper according to our official guidelines. This is one of the highlights of publishing with Global Journals—authors should not be concerned about the formatting of their paper. Global Journals accepts articles and manuscripts in every major language, be it Spanish, Chinese, Japanese, Portuguese, Russian, French, German, Dutch, Italian, Greek, or any other national language, but the title, subtitle, and abstract should be in English. This will facilitate indexing and the pre-peer review process.

The following is the official style and template developed for publication of a research paper. Authors are not required to follow this style during the submission of the paper. It is just for reference purposes.

Manuscript Style Instruction (Optional)

- Microsoft Word Document Setting Instructions.
- Font type of all text should be Swis721 Lt BT.
- Page size: 8.27" x 11'", left margin: 0.65, right margin: 0.65, bottom margin: 0.75.
- Paper title should be in one column of font size 24.
- Author name in font size of 11 in one column.
- Abstract: font size 9 with the word "Abstract" in bold italics.
- Main text: font size 10 with two justified columns.
- Two columns with equal column width of 3.38 and spacing of 0.2.
- First character must be three lines drop-capped.
- The paragraph before spacing of 1 pt and after of 0 pt.
- Line spacing of 1 pt.
- Large images must be in one column.
- The names of first main headings (Heading 1) must be in Roman font, capital letters, and font size of 10.
- The names of second main headings (Heading 2) must not include numbers and must be in italics with a font size of 10.

Structure and Format of Manuscript

The recommended size of an original research paper is under 15,000 words and review papers under 7,000 words. Research articles should be less than 10,000 words. Research papers are usually longer than review papers. Review papers are reports of significant research (typically less than 7,000 words, including tables, figures, and references)

A research paper must include:

a) A title which should be relevant to the theme of the paper.
b) A summary, known as an abstract (less than 150 words), containing the major results and conclusions.
c) Up to 10 keywords that precisely identify the paper's subject, purpose, and focus.
d) An introduction, giving fundamental background objectives.
e) Resources and techniques with sufficient complete experimental details (wherever possible by reference) to permit repetition, sources of information must be given, and numerical methods must be specified by reference.
f) Results which should be presented concisely by well-designed tables and figures.
g) Suitable statistical data should also be given.
h) All data must have been gathered with attention to numerical detail in the planning stage.

Design has been recognized to be essential to experiments for a considerable time, and the editor has decided that any paper that appears not to have adequate numerical treatments of the data will be returned unrefereed.

i) Discussion should cover implications and consequences and not just recapitulate the results; conclusions should also be summarized.
j) There should be brief acknowledgments.
k) There ought to be references in the conventional format. Global Journals recommends APA format.

Authors should carefully consider the preparation of papers to ensure that they communicate effectively. Papers are much more likely to be accepted if they are carefully designed and laid out, contain few or no errors, are summarizing, and follow instructions. They will also be published with much fewer delays than those that require much technical and editorial correction.

The Editorial Board reserves the right to make literary corrections and suggestions to improve brevity.

Format Structure

It is necessary that authors take care in submitting a manuscript that is written in simple language and adheres to published guidelines.

All manuscripts submitted to Global Journals should include:

Title

The title page must carry an informative title that reflects the content, a running title (less than 45 characters together with spaces), names of the authors and co-authors, and the place(s) where the work was carried out.

Author details

The full postal address of any related author(s) must be specified.

Abstract

The abstract is the foundation of the research paper. It should be clear and concise and must contain the objective of the paper and inferences drawn. It is advised to not include big mathematical equations or complicated jargon.

Many researchers searching for information online will use search engines such as Google, Yahoo or others. By optimizing your paper for search engines, you will amplify the chance of someone finding it. In turn, this will make it more likely to be viewed and cited in further works. Global Journals has compiled these guidelines to facilitate you to maximize the web-friendliness of the most public part of your paper.

Keywords

A major lynchpin of research work for the writing of research papers is the keyword search, which one will employ to find both library and internet resources. Up to eleven keywords or very brief phrases have to be given to help data retrieval, mining, and indexing.

One must be persistent and creative in using keywords. An effective keyword search requires a strategy: planning of a list of possible keywords and phrases to try.

Choice of the main keywords is the first tool of writing a research paper. Research paper writing is an art. Keyword search should be as strategic as possible.

One should start brainstorming lists of potential keywords before even beginning searching. Think about the most important concepts related to research work. Ask, “What words would a source have to include to be truly valuable in a research paper?” Then consider synonyms for the important words.

It may take the discovery of only one important paper to steer in the right keyword direction because, in most databases, the keywords under which a research paper is abstracted are listed with the paper.

Numerical Methods

Numerical methods used should be transparent and, where appropriate, supported by references.

Abbreviations

Authors must list all the abbreviations used in the paper at the end of the paper or in a separate table before using them.

Formulas and equations

Authors are advised to submit any mathematical equation using either MathJax, KaTeX, or LaTeX, or in a very high-quality image.

Tables, Figures, and Figure Legends

Tables: Tables should be cautiously designed, uncrowned, and include only essential data. Each must have an Arabic number, e.g., Table 4, a self-explanatory caption, and be on a separate sheet. Authors must submit tables in an editable format and not as images. References to these tables (if any) must be mentioned accurately.

Figures

Figures are supposed to be submitted as separate files. Always include a citation in the text for each figure using Arabic numbers, e.g., Fig. 4. Artwork must be submitted online in vector electronic form or by emailing it.

Preparation of Eletronic Figures for Publication

Although low-quality images are sufficient for review purposes, print publication requires high-quality images to prevent the final product being blurred or fuzzy. Submit (possibly by e-mail) EPS (line art) or TIFF (halftone/ photographs) files only. MS PowerPoint and Word Graphics are unsuitable for printed pictures. Avoid using pixel-oriented software. Scans (TIFF only) should have a resolution of at least 350 dpi (halftone) or 700 to 1100 dpi (line drawings). Please give the data for figures in black and white or submit a Color Work Agreement form. EPS files must be saved with fonts embedded (and with a TIFF preview, if possible).

For scanned images, the scanning resolution at final image size ought to be as follows to ensure good reproduction: line art: >650 dpi; halftones (including gel photographs): >350 dpi; figures containing both halftone and line images: >650 dpi.

Color charges: Authors are advised to pay the full cost for the reproduction of their color artwork. Hence, please note that if there is color artwork in your manuscript when it is accepted for publication, we would require you to complete and return a Color Work Agreement form before your paper can be published. Also, you can email your editor to remove the color fee after acceptance of the paper.

Tips for writing a good quality Medical Research Paper

1. Choosing the topic: In most cases, the topic is selected by the interests of the author, but it can also be suggested by the guides. You can have several topics, and then judge which you are most comfortable with. This may be done by asking several questions of yourself, like "Will I be able to carry out a search in this area? Will I find all necessary resources to accomplish the search? Will I be able to find all information in this field area?" If the answer to this type of question is "yes," then you ought to choose that topic. In most cases, you may have to conduct surveys and visit several places. Also, you might have to do a lot of work to find all the rises and falls of the various data on that subject. Sometimes, detailed information plays a vital role, instead of short information. Evaluators are human: The first thing to remember is that evaluators are also human beings. They are not only meant for rejecting a paper. They are here to evaluate your paper. So present your best aspect.

2. Think like evaluators: If you are in confusion or getting demotivated because your paper may not be accepted by the evaluators, then think, and try to evaluate your paper like an evaluator. Try to understand what an evaluator wants in your research paper, and you will automatically have your answer. Make blueprints of paper: The outline is the plan or framework that will help you to arrange your thoughts. It will make your paper logical. But remember that all points of your outline must be related to the topic you have chosen.

3. Ask your guides: If you are having any difficulty with your research, then do not hesitate to share your difficulty with your guide (if you have one). They will surely help you out and resolve your doubts. If you can't clarify what exactly you require for your work, then ask your supervisor to help you with an alternative. He or she might also provide you with a list of essential readings.

4. Use of computer is recommended: As you are doing research in the field of medical research then this point is quite obvious. Use right software: Always use good quality software packages. If you are not capable of judging good software, then you can lose the quality of your paper unknowingly. There are various programs available to help you which you can get through the internet.

5. Use the internet for help: An excellent start for your paper is using Google. It is a wondrous search engine, where you can have your doubts resolved. You may also read some answers for the frequent question of how to write your research paper or find a model research paper. You can download books from the internet. If you have all the required books, place importance on reading, selecting, and analyzing the specified information. Then sketch out your research paper. Use big pictures: You may use encyclopedias like Wikipedia to get pictures with the best resolution. At Global Journals, you should strictly follow here.

6. Bookmarks are useful: When you read any book or magazine, you generally use bookmarks, right? It is a good habit which helps to not lose your continuity. You should always use bookmarks while searching on the internet also, which will make your search easier.

7. Revise what you wrote: When you write anything, always read it, summarize it, and then finalize it.

8. Make every effort: Make every effort to mention what you are going to write in your paper. That means always have a good start. Try to mention everything in the introduction—what is the need for a particular research paper. Polish your work with good writing skills and always give an evaluator what he wants. Make backups: When you are going to do any important thing like making a research paper, you should always have backup copies of it either on your computer or on paper. This protects you from losing any portion of your important data.

9. Produce good diagrams of your own: Always try to include good charts or diagrams in your paper to improve quality. Using several unnecessary diagrams will degrade the quality of your paper by creating a hodgepodge. So always try to include diagrams which were made by you to improve the readability of your paper. Use of direct quotes: When you do research relevant to literature, history, or current affairs, then use of quotes becomes essential, but if the study is relevant to science, use of quotes is not preferable.

10. Use proper verb tense: Use proper verb tenses in your paper. Use past tense to present those events that have happened. Use present tense to indicate events that are going on. Use future tense to indicate events that will happen in the future. Use of wrong tenses will confuse the evaluator. Avoid sentences that are incomplete.

11. Pick a good study spot: Always try to pick a spot for your research which is quiet. Not every spot is good for studying.

12. Know what you know: Always try to know what you know by making objectives, otherwise you will be confused and unable to achieve your target.

13. Use good grammar: Always use good grammar and words that will have a positive impact on the evaluator; use of good vocabulary does not mean using tough words which the evaluator has to find in a dictionary. Do not fragment sentences. Eliminate one-word sentences. Do not ever use a big word when a smaller one would suffice.

Verbs have to be in agreement with their subjects. In a research paper, do not start sentences with conjunctions or finish them with prepositions. When writing formally, it is advisable to never split an infinitive because someone will (wrongly) complain. Avoid clichés like a disease. Always shun irritating alliteration. Use language which is simple and straightforward. Put together a neat summary.

14. Arrangement of information: Each section of the main body should start with an opening sentence, and there should be a changeover at the end of the section. Give only valid and powerful arguments for your topic. You may also maintain your arguments with records.

15. Never start at the last minute: Always allow enough time for research work. Leaving everything to the last minute will degrade your paper and spoil your work.

16. Multitasking in research is not good: Doing several things at the same time is a bad habit in the case of research activity. Research is an area where everything has a particular time slot. Divide your research work into parts, and do a particular part in a particular time slot.

17. Never copy others' work: Never copy others' work and give it your name because if the evaluator has seen it anywhere, you will be in trouble. Take proper rest and food: No matter how many hours you spend on your research activity, if you are not taking care of your health, then all your efforts will have been in vain. For quality research, take proper rest and food.

18. Go to seminars: Attend seminars if the topic is relevant to your research area. Utilize all your resources.

19. Refresh your mind after intervals: Try to give your mind a rest by listening to soft music or sleeping in intervals. This will also improve your memory. Acquire colleagues: Always try to acquire colleagues. No matter how sharp you are, if you acquire colleagues, they can give you ideas which will be helpful to your research.

20. Think technically: Always think technically. If anything happens, search for its reasons, benefits, and demerits. Think and then print: When you go to print your paper, check that tables are not split, headings are not detached from their descriptions, and page sequence is maintained.

21. Adding unnecessary information: Do not add unnecessary information like "I have used MS Excel to draw graphs." Irrelevant and inappropriate material is superfluous. Foreign terminology and phrases are not apropos. One should never take a broad view. Analogy is like feathers on a snake. Use words properly, regardless of how others use them. Remove quotations. Puns are for kids, not grunt readers. Never oversimplify: When adding material to your research paper, never go for oversimplification; this will definitely irritate the evaluator. Be specific. Never use rhythmic redundancies. Contractions shouldn't be used in a research paper. Comparisons are as terrible as clichés. Give up ampersands, abbreviations, and so on. Remove commas that are not necessary. Parenthetical words should be between brackets or commas. Understatement is always the best way to put forward earth-shaking thoughts. Give a detailed literary review.

22. Report concluded results: Use concluded results. From raw data, filter the results, and then conclude your studies based on measurements and observations taken. An appropriate number of decimal places should be used. Parenthetical remarks are prohibited here. Proofread carefully at the final stage. At the end, give an outline to your arguments. Spot perspectives of further study of the subject. Justify your conclusion at the bottom sufficiently, which will probably include examples.

23. Upon conclusion: Once you have concluded your research, the next most important step is to present your findings. Presentation is extremely important as it is the definite medium though which your research is going to be in print for the rest of the crowd. Care should be taken to categorize your thoughts well and present them in a logical and neat manner. A good quality research paper format is essential because it serves to highlight your research paper and bring to light all necessary aspects of your research.

Informal Guidelines of Research Paper Writing

Key points to remember:

- Submit all work in its final form.
- Write your paper in the form which is presented in the guidelines using the template.
- Please note the criteria peer reviewers will use for grading the final paper.

Final points:

One purpose of organizing a research paper is to let people interpret your efforts selectively. The journal requires the following sections, submitted in the order listed, with each section starting on a new page:

The introduction: This will be compiled from reference matter and reflect the design processes or outline of basis that directed you to make a study. As you carry out the process of study, the method and process section will be constructed like that. The results segment will show related statistics in nearly sequential order and direct reviewers to similar intellectual paths throughout the data that you gathered to carry out your study.

The discussion section:

This will provide understanding of the data and projections as to the implications of the results. The use of good quality references throughout the paper will give the effort trustworthiness by representing an alertness to prior workings.

Writing a research paper is not an easy job, no matter how trouble-free the actual research or concept. Practice, excellent preparation, and controlled record-keeping are the only means to make straightforward progression.

General style:

Specific editorial column necessities for compliance of a manuscript will always take over from directions in these general guidelines.

To make a paper clear: Adhere to recommended page limits.

Mistakes to avoid:

- Insertion of a title at the foot of a page with subsequent text on the next page.
- Separating a table, chart, or figure—confine each to a single page.
- Submitting a manuscript with pages out of sequence.
- In every section of your document, use standard writing style, including articles ("a" and "the").
- Keep paying attention to the topic of the paper.
- Use paragraphs to split each significant point (excluding the abstract).
- Align the primary line of each section.
- Present your points in sound order.
- Use present tense to report well-accepted matters.
- Use past tense to describe specific results.
- Do not use familiar wording; don't address the reviewer directly. Don't use slang or superlatives.
- Avoid use of extra pictures—include only those figures essential to presenting results.

Title page:

Choose a revealing title. It should be short and include the name(s) and address(es) of all authors. It should not have acronyms or abbreviations or exceed two printed lines.

Abstract: This summary should be two hundred words or less. It should clearly and briefly explain the key findings reported in the manuscript and must have precise statistics. It should not have acronyms or abbreviations. It should be logical in itself. Do not cite references at this point.

An abstract is a brief, distinct paragraph summary of finished work or work in development. In a minute or less, a reviewer can be taught the foundation behind the study, common approaches to the problem, relevant results, and significant conclusions or new questions.

Write your summary when your paper is completed because how can you write the summary of anything which is not yet written? Wealth of terminology is very essential in abstract. Use comprehensive sentences, and do not sacrifice readability for brevity; you can maintain it succinctly by phrasing sentences so that they provide more than a lone rationale. The author can at this moment go straight to shortening the outcome. Sum up the study with the subsequent elements in any summary. Try to limit the initial two items to no more than one line each.

Reason for writing the article—theory, overall issue, purpose.

- Fundamental goal.
- To-the-point depiction of the research.
- Consequences, including definite statistics—if the consequences are quantitative in nature, account for this; results of any numerical analysis should be reported. Significant conclusions or questions that emerge from the research.

Approach:

- Single section and succinct.
- An outline of the job done is always written in past tense.
- Concentrate on shortening results—limit background information to a verdict or two.
- Exact spelling, clarity of sentences and phrases, and appropriate reporting of quantities (proper units, important statistics) are just as significant in an abstract as they are anywhere else.

Introduction:

The introduction should "introduce" the manuscript. The reviewer should be presented with sufficient background information to be capable of comprehending and calculating the purpose of your study without having to refer to other works. The basis for the study should be offered. Give the most important references, but avoid making a comprehensive appraisal of the topic. Describe the problem visibly. If the problem is not acknowledged in a logical, reasonable way, the reviewer will give no attention to your results. Speak in common terms about techniques used to explain the problem, if needed, but do not present any particulars about the protocols here.

The following approach can create a valuable beginning:

- Explain the value (significance) of the study.
- Defend the model—why did you employ this particular system or method? What is its compensation? Remark upon its appropriateness from an abstract point of view as well as pointing out sensible reasons for using it.
- Present a justification. State your particular theory(-ies) or aim(s), and describe the logic that led you to choose them.
- Briefly explain the study's tentative purpose and how it meets the declared objectives.

Approach:

Use past tense except for when referring to recognized facts. After all, the manuscript will be submitted after the entire job is done. Sort out your thoughts; manufacture one key point for every section. If you make the four points listed above, you will need at least four paragraphs. Present surrounding information only when it is necessary to support a situation. The reviewer does not desire to read everything you know about a topic. Shape the theory specifically—do not take a broad view.

As always, give awareness to spelling, simplicity, and correctness of sentences and phrases.

Procedures (methods and materials):

This part is supposed to be the easiest to carve if you have good skills. A soundly written procedures segment allows a capable scientist to replicate your results. Present precise information about your supplies. The suppliers and clarity of reagents can be helpful bits of information. Present methods in sequential order, but linked methodologies can be grouped as a segment. Be concise when relating the protocols. Attempt to give the least amount of information that would permit another capable scientist to replicate your outcome, but be cautious that vital information is integrated. The use of subheadings is suggested and ought to be synchronized with the results section.

When a technique is used that has been well-described in another section, mention the specific item describing the way, but draw the basic principle while stating the situation. The purpose is to show all particular resources and broad procedures so that another person may use some or all of the methods in one more study or referee the scientific value of your work. It is not to be a step-by-step report of the whole thing you did, nor is a methods section a set of orders.

Materials:

Materials may be reported in part of a section or else they may be recognized along with your measures.

Methods:

- Report the method and not the particulars of each process that engaged the same methodology.
- Describe the method entirely.
- To be succinct, present methods under headings dedicated to specific dealings or groups of measures.
- Simplify—detail how procedures were completed, not how they were performed on a particular day.
- If well-known procedures were used, account for the procedure by name, possibly with a reference, and that's all.

Approach:

It is embarrassing to use vigorous voice when documenting methods without using first person, which would focus the reviewer's interest on the researcher rather than the job. As a result, when writing up the methods, most authors use third person passive voice.

Use standard style in this and every other part of the paper—avoid familiar lists, and use full sentences.

What to keep away from:

- Resources and methods are not a set of information.
- Skip all descriptive information and surroundings—save it for the argument.
- Leave out information that is immaterial to a third party.

Results:

The principle of a results segment is to present and demonstrate your conclusion. Create this part as entirely objective details of the outcome, and save all understanding for the discussion.

The page length of this segment is set by the sum and types of data to be reported. Use statistics and tables, if suitable, to present consequences most efficiently.

You must clearly differentiate material which would usually be incorporated in a study editorial from any unprocessed data or additional appendix matter that would not be available. In fact, such matters should not be submitted at all except if requested by the instructor.

Content:

- Sum up your conclusions in text and demonstrate them, if suitable, with figures and tables.
- In the manuscript, explain each of your consequences, and point the reader to remarks that are most appropriate.
- Present a background, such as by describing the question that was addressed by creation of an exacting study.
- Explain results of control experiments and give remarks that are not accessible in a prescribed figure or table, if appropriate.
- Examine your data, then prepare the analyzed (transformed) data in the form of a figure (graph), table, or manuscript.

What to stay away from:

- Do not discuss or infer your outcome, report surrounding information, or try to explain anything.
- Do not include raw data or intermediate calculations in a research manuscript.
- Do not present similar data more than once.
- A manuscript should complement any figures or tables, not duplicate information.
- Never confuse figures with tables—there is a difference.

Approach:

As always, use past tense when you submit your results, and put the whole thing in a reasonable order.

Put figures and tables, appropriately numbered, in order at the end of the report.

If you desire, you may place your figures and tables properly within the text of your results section.

Figures and tables:

If you put figures and tables at the end of some details, make certain that they are visibly distinguished from any attached appendix materials, such as raw facts. Whatever the position, each table must be titled, numbered one after the other, and include a heading. All figures and tables must be divided from the text.

Discussion:

The discussion is expected to be the trickiest segment to write. A lot of papers submitted to the journal are discarded based on problems with the discussion. There is no rule for how long an argument should be.

Position your understanding of the outcome visibly to lead the reviewer through your conclusions, and then finish the paper with a summing up of the implications of the study. The purpose here is to offer an understanding of your results and support all of your conclusions, using facts from your research and generally accepted information, if suitable. The implication of results should be fully described.

Infer your data in the conversation in suitable depth. This means that when you clarify an observable fact, you must explain mechanisms that may account for the observation. If your results vary from your prospect, make clear why that may have happened. If your results agree, then explain the theory that the proof supported. It is never suitable to just state that the data approved the prospect, and let it drop at that. Make a decision as to whether each premise is supported or discarded or if you cannot make a conclusion with assurance. Do not just dismiss a study or part of a study as "uncertain."

Research papers are not acknowledged if the work is imperfect. Draw what conclusions you can based upon the results that you have, and take care of the study as a finished work.

- You may propose future guidelines, such as how an experiment might be personalized to accomplish a new idea.
- Give details of all of your remarks as much as possible, focusing on mechanisms.
- Make a decision as to whether the tentative design sufficiently addressed the theory and whether or not it was correctly restricted. Try to present substitute explanations if they are sensible alternatives.
- One piece of research will not counter an overall question, so maintain the large picture in mind. Where do you go next? The best studies unlock new avenues of study. What questions remain?
- Recommendations for detailed papers will offer supplementary suggestions.

Approach:

When you refer to information, differentiate data generated by your own studies from other available information. Present work done by specific persons (including you) in past tense.

Describe generally acknowledged facts and main beliefs in present tense.

The Administration Rules

Administration Rules to Be Strictly Followed before Submitting Your Research Paper to Global Journals Inc.

Please read the following rules and regulations carefully before submitting your research paper to Global Journals Inc. to avoid rejection.

Segment draft and final research paper: You have to strictly follow the template of a research paper, failing which your paper may get rejected. You are expected to write each part of the paper wholly on your own. The peer reviewers need to identify your own perspective of the concepts in your own terms. Please do not extract straight from any other source, and do not rephrase someone else's analysis. Do not allow anyone else to proofread your manuscript.

Written material: You may discuss this with your guides and key sources. Do not copy anyone else's paper, even if this is only imitation, otherwise it will be rejected on the grounds of plagiarism, which is illegal. Various methods to avoid plagiarism are strictly applied by us to every paper, and, if found guilty, you may be blacklisted, which could affect your career adversely. To guard yourself and others from possible illegal use, please do not permit anyone to use or even read your paper and file.

Criterion for Grading a Research Paper (Compilation) by Global Journals

Please note that following table is only a Grading of "Paper Compilation" and not on "Performed/Stated Research" whose grading solely depends on Individual Assigned Peer Reviewer and Editorial Board Member. These can be available only on request and after decision of Paper. This report will be the property of Global Journals.

Topics	Grades		
	A-B	**C-D**	**E-F**
Abstract	Clear and concise with appropriate content, Correct format. 200 words or below	Unclear summary and no specific data, Incorrect form Above 200 words	No specific data with ambiguous information Above 250 words
Introduction	Containing all background details with clear goal and appropriate details, flow specification, no grammar and spelling mistake, well organized sentence and paragraph, reference cited	Unclear and confusing data, appropriate format, grammar and spelling errors with unorganized matter	Out of place depth and content, hazy format
Methods and Procedures	Clear and to the point with well arranged paragraph, precision and accuracy of facts and figures, well organized subheads	Difficult to comprehend with embarrassed text, too much explanation but completed	Incorrect and unorganized structure with hazy meaning
Result	Well organized, Clear and specific, Correct units with precision, correct data, well structuring of paragraph, no grammar and spelling mistake	Complete and embarrassed text, difficult to comprehend	Irregular format with wrong facts and figures
Discussion	Well organized, meaningful specification, sound conclusion, logical and concise explanation, highly structured paragraph reference cited	Wordy, unclear conclusion, spurious	Conclusion is not cited, unorganized, difficult to comprehend
References	Complete and correct format, well organized	Beside the point, Incomplete	Wrong format and structuring

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ISSN 9755896